State of Hawaii
DEPARTMENT OF LAND AND NATURAL RESOURCES
Division of Aquatic Resources
Honolulu, Hawaii 96813

April 28, 2006

Board of Land
and Natural Resources
Honolulu, Hawaii

THE DIVISION OF AQUATIC RESOURCES REQUESTS BOARD OF LAND AND NATURAL RESOURCES (BLNR) AUTHORIZATION/APPROVAL TO ISSUE TWO (2) NORTHWESTERN HAWAIIAN ISLANDS (NWHI) RESEARCH, MONITORING AND EDUCATION PERMITS TO: 1) DR MICHAEL RAPPÉ, AND 2) DR. RUTH GATES, BOTH OF THE HAWAII INSTITUTE OF MARINE BIOLOGY, FOR THE NON-LETHAL SAMPLING OF CORALS TO IDENTIFY BIOLOGICAL INDICATORS OF CORAL DISEASE AND/OR BLEACHING SUSCEPTIBILITY

Submitted herewith for your authorization and approval is a request for issuance of two (2) Research, Monitoring and Education permits to Drs. Michael Rappé and Ruth Gates of the Hawaii Institute of Marine Biology, University of Hawaii. These permits, described below, will allow activity to occur in the NWHI State marine Refuge (0-3 miles) waters surrounding Nihoa Island, French Frigate Shoals, and Gardner Pinnacles. The activities covered under this permit will occur from May 18 to June 11, 2006, from the support vessel Hi'ialakai. Ship details are provided with Item F-4.

The proposed activities (below) are consistent with and support the purposes of the Refuge, primarily to better understand and manage the resources within the marine refuge. Understanding the causes of coral disease and developing biological indicators for disease and bleaching are of value to State resource managers. Such information will allow managers to identify specific coral populations that are particularly fragile, or at risk for disease outbreaks or bleaching, and propose appropriate management strategies.

1. RESEARCH, MONITORING AND EDUCATION PERMIT TO RAPPÉ:

Microbial pathogens have been identified as causative agents for a variety of diseases that have devastated coral reefs world wide; however, healthy corals are also known to host diverse and unique communities of microorganisms. Determining disease etiologies is difficult because little is known about the microbial communities of healthy corals. The purpose of Rappé's study is to identify the diversity of microbes associated with different species of healthy and health-compromised corals in the NWHI, and to determine the extent to which invasive microbial pathogens are present on NWHI reefs. The ultimate goal of his study is to be able to assess coral health and predict disease susceptibility for individual corals. Rappé requires non-lethal biopsies of 6mm^2, the size of a small fish bite or pencil eraser, and will be sub-sampling from coral samples to be shared by the HIMB collaboration (Aeby, Gates, Karl, Rappé, and Toonen – see attached Sampling Table).

2. RESEARCH, MONITORING AND EDUCATION PERMIT TO GATES:

The goal of Gates' research is to identify robust biological indicators of coral disease and/or bleaching susceptibility. Specifically, she will characterize the diversity of symbiotic dinoflagellates harbored by corals and examine morphological traits such as polyp size, colony size, and colony morphotype in healthy, diseased and/or bleached corals to identify biological traits that correlate with health state. She will take samples that will be analyzed at the Hawaii Institute of Marine Biology with molecular and other laboratory techniques. This research is directly relevant to management in: 1) identifying areas of the State Refuge that have a high incidence of disease- and/or bleaching-susceptible individuals; 2) tracking the spread and prevalence of symbiont types that are associated with disease and/or bleaching susceptibility, and 3) providing a detailed baseline against which future monitoring activities can be compared.

REVIEW PROCESS:

The permits were received by the Division of Aquatic Resources on 1) March 6, 2006 and 2) March 8, 2006. They were sent out for review and comment to the following scientific entities: Division of Aquatic Resources staff (5), Division of Forestry and Wildlife, Northwest Hawaiian Islands Reserve, and the United States Fish and Wildlife Service. Native Hawaiians from the Office of Hawaiian Affairs and Kaho'olawe Island Reserve Commission were also consulted.

Comments received from the Scientific Community (DAR) on Rappé's Permit are summarized as follows:

1) Concern was expressed that Rappé might want to transport live disease cultures aboard ship
2) There was concern that appropriate sampling and diving gear protocols should be developed and enforced so as not to spread any coral disease among sites in the NWHI
3) Concern was expressed about open-ended sampling language (e.g., "at least 5 colonies") in the original proposal

Comments received from the Scientific Community (DAR) on Gates' Permit are summarized as follows:

1) One reviewer complimented the proposal, small sample size and connectivity with other projects
2) Concern was expressed that some of the species to be sampled are branching forms and provide essential fish habitat
3) Concern was expressed over the sampling of rare or uncommon species
4) There was concern that appropriate sampling and diving gear protocols should be developed and enforced so as not to spread any coral disease among sites in the NWHI

Comments received from a Native Hawaiian on both the Research, Education and Monitoring Permits are summarized as follows:

1) There was concern for native Hawaiian intellectual property rights for new discoveries and the protection of the resources for their potential product developments.

RESPONSE:

A meeting of DAR staff and HIMB researchers was held on 12 April 2006 to address concerns, and a synopsis of the response to concerns raised is as follows:

Rappé:

1) Rappé has no desire to transport disease, and does not ask for permission to do so. All of his samples will be killed immediately by freezing aboard ship.

2) Protocols are already in place for disinfection of sampling and diving gear between sites in the NWHI. All gear is soaked in 10% bleach solution to kill any microorganisms.

3) Samples will be limited to the species collection table provided by HIMB researchers. Rappé's samples are about the size of a fish bite, and he is interested in common and widespread species. Coral samples are to be taken in coordination with other HIMB personnel: the stated sample sizes and numbers are to be shared by Toonen, Aeby, Karl, Rappé and Gates (see separate permit applications).

4) The Guidelines for Submitting Permit Applications stipulates that, for all permits, the activity must be non-commercial and will not involve the sale of any organism, byproduct, or material collected. Furthermore, the Guidelines state that resources and samples are a public trust, and are not to be used for sale, patent, bioassay, or bio-prospecting, or for obtaining patents or intellectual property rights. This condition will be added to the Permit Terms and Conditions for this, and all future permits. This should address the concerns raised by the Native Hawaiian reviewer.

Gates:

1) Gates replied that utmost care is taken to prevent damage to branching forms, and very small (2 cm^2) samples are required for her work. Furthermore, she only proposes to take 5 biopsies of rare or uncommon species per site (included on the HIMB Coral Sampling Table (attached).

2) Protocols are already in place for disinfection of sampling and diving gear between sites in the NWHI. Gates' samples are killed by freezing aboard the ship. All gear is soaked in 10% bleach solution between sites to kill any microorganisms and eliminate the possibility of disease transmission.

3) The Guidelines for Submitting Permit Applications stipulates that, for all permits, the activity must be non-commercial and will not involve the sale of any organism, byproduct, or material collected. Furthermore, the Guidelines state that

resources and samples are a public trust, and are not to be used for sale, patent, bioassay, or bio-prospecting, or for obtaining patents or intellectual property rights. This condition will be added to the Permit Terms and Conditions for this, and all future permits. This should address the concerns raised by the Native Hawaiian reviewer.

AMENDMENTS REQUESTED SUBSEQUENT TO APPLICATION SUBMISSION:

Rappé requested via email on April 18, 2006 that Anderson Mayfield be added to his permit application as a sub-permittee

FINAL STAFF RECOMMENDATIONS:

1) Allow Rappé and Gates to take non-lethal samples of corals, not to exceed the numbers specified in the HIMB Coral Sampling Table (attached). Coral samples are to be taken in coordination with other HIMB personnel: the stated sample sizes and numbers are to be shared by Toonen, Aeby, Karl, Rappé and Gates (see separate permit applications).
2) Allow the addition of personnel to Rappé's permit

RECOMMENDATION:

"That the Board authorize and approve, with stated conditions, 1) a Research, Monitoring and Education Permit to Dr. Michael Rappé of the Hawaii Institute of Marine Biology, and 2) a Research, Monitoring and Education Permit to Dr. Ruth Gates of the Hawaii Institute of Marine Biology, for activities and access within the State waters of the NWHI."

Respectfully submitted,

DAN POLHEMUS
Administrator

APPROVED FOR SUBMITTAL

PETER T. YOUNG
Chairperson

APPENDIX 1

State of Hawai'i
DLNR
Northwestern Hawaiian Islands State Marine Refuge
Permit Application Form

For Office Use Only
Permit No:
Expiration date:
Date Appl. Received: 3/6/06
Appl. Fee received: N/A
NWHI Permit Review Committee date:
Board Hearing date:
Post to web date:

Type of Permit

☑ I am applying for a **Research, Monitoring & Education** permit. (Complete and mail Application)
- ☑ This application is for a NEW project in the State Marine Refuge.
- ☐ This application is for an ANNUAL RENEWAL of a previously permitted project in the State Marine Refuge.

☐ I am applying for a permit for a **Native Hawaiian** permit. (Complete and mail Application)

- ☐ This application is for a NEW project in the State Marine Refuge.
- ☐ This application is for an ANNUAL RENEWAL of a previously permitted project in the State Marine Refuge.

☑ I am applying for a **Special Activity** permit. (Complete and mail Application)

- ☑ This application is for a NEW project in the State Marine Refuge.
- ☑ This application is for an ANNUAL RENEWAL of a previously permitted project in the State Marine Refuge.

Briefly describe **Special permit** activity:

When will the NWHI activity take place?

☑ **Summer** (May-July of 2006 (year)
Note: Permit request must be received before February 1st
Specific dates of expedition _____

☐ **Fall** (August-November) of ____ (year)
Note: Permit request must be received before May 1st
Specific dates of expedition _____

☐ **Other**

NOTE: INCOMPLETE APPLICATIONS WILL NOT BE ACCEPTED

Please Send Permit Applications to:

NWHI State Marine Refuge Permit Coordinator
State of Hawai'i
Department of Land and Natural Resources
Division of Aquatic Resources
1151 Punchbowl Street, Room 330
Honolulu, Hawai'i 96813

NWHI State Marine Refuge Permit Application
See Appendix 2 for Application Instructions

Section A – Applicant Information

1. Project Leader (attach Project Leader's CV or resume) ☑ CV attached Rappe, Michael S. Name: Last, First, Middle Initial	Assistant Research Professor Title
2. Mailing Address (Street/PO Box, City, State, Zip) School of Ocean & Earth Science & Technology University of Hawaii Hawaii Institute of Marine Biology	Telephone (808) 236-7464 Fax (808) 236-7443 Email Address rappe@hawaii.edu
3. Affiliation (Institution/Agency/Organization) SOEST, Hawaii Institute of Marine Biology, University of Hawaii	For graduate students, Major Professor 's Name & Telephone
4. Sub-Permittee/Assistant Names, Affiliations, and Contact Information ☐ CV or resume attached Jennifer L. Salerno, Ph.D. Graduate Student Department of Zoology, University of Hawaii at Manoa	
5. Project Title Assessing diversity of microbes associated with corals in the NWHI	
6. Applicant Signature 	7. Date (mm/dd/yyyy) 03/08/2006

Section B: Project Information

8. **(a) Project Location**

☑ NWHI State Marine Refuge (0-3 miles) waters surrounding:

- ☑ Nihoa Island
- ☐ Necker Island (Mokumanamana)
- ☑ French Frigate Shoals
- ☐ Laysan
- ☐ Maro
- ☑ Gardner Pinnacles
- ☐ Lisianski Island, Neva Shoal
- ☐ Pearl and Hermes Atoll
- ☐ Kure Atoll, State Wildlife Refuge
- ☐ Other NWHI location

Describe project location (include names, GPS coordinates, habitats, depths and attach maps, etc. as appropriate).

(b) check all actions to be authorized:

☑ Enter the NWHI Marine Refuge waters

☑ Take (harvest) ☐ Possess ☑ Transport (☑ Inter-island ☐ Out-of-state)

☐ Catch ☐ Kill ☐ Disturb ☑ Observe

☑ Anchor ☐ Land (go ashore) ☐ Archaeological research

☐ Interactions with Sea Turtles or Monk Seals ☐ Interactions with Seabirds

☑ Interactions with Live Coral, Ark Shells or Pearl Oysters

☐ Interactions with Jacks, Grouper or Sharks

☐ Conduct Native Hawaiian religious and/or cultural activities

☑ Other activities non-lethal biopsies taken from corals and frozen

(c) Collection of specimens – collecting activities (would apply to any activity):

Organisms or objects (List of species, if applicable, add additional sheets if necessary):

Common name	**Scientific name**	**No. & size of specimens**	**Collection Location(s)**

Please see question 8 of the attached Appendix.

(d) What will be done with the specimens after the project has ended?

Some archival coral samples will be stored frozen, however, the majority of samples will be completely consumed during processing for molecular analysis.
Please refer to question 10 Procedures on the attached Appendix for additional information.

(e) Will the organisms be kept alive after collection? ☐ yes ☑ no

- Specific site/location _____
- Is it an open or closed system? ☐ open ☐ closed
- Is there an outfall? ☐ yes ☐ no
- Will these organisms be housed with other organisms? If so, what are the other organisms?

(Please attach additional documentation as needed to complete the questions listed below)

9. Purpose/Need/Scope:

- Please refer to question 9 of attached Appendix.

Describe how your proposed activities will help provide information or resources to fulfill the State Marine Refuge purpose and to reach the Refuge goals and objectives.

- Please refer to question 9 of attached Appendix.

- Describe context of this activity, include history of the science for these questions and background.

  Please refer to question 9 of attached Appendix.

- Explain the need for this activity and how it will help to enhance survival or recovery of refuge wildlife and habitats.

  Please refer to question 9 of attached Appendix.

- Describe how your proposed project can help to better manage the State Marine Refuge.

  Please refer to question 9 of attached Appendix.

10. Procedures (include equipment/materials)

Please refer to question 10 of attached Appendix.

11. Funding sources (attach copies budget & funding sources).

Project: HIMB-NWHI Coral Reef Research Partnership
Reference number: NMSP MOA 2005-008/66882

12. List all literature cited in this application as well as all other publications relevant to the proposed project.

Please refer to question 12 of attached Appendix.

13. What types of insurance do you have in place? (attach documentation)

☑ Wreck Removal

☑ Pollution

14. What certifications/inspections do you have scheduled for your vessel? (attach documentation)

☑ Rat free ☐ tender vessel ☐ gear/equipment

☑ Hull inspection ☑ ballast water

15. Other permits (list and attach documentation of all other required Federal or State permits).

Applications for USFWS Special Use and a NWHI Coral Reef Ecosystem Reserve Research Permit are in review.

16. Project's relationship to other research projects within the NWHI State Marine Refuge, National Wildlife Refuge, NWHI Coral Reef Ecosystem Reserve, or elsewhere.

Please refer to question 16 of the attached Appendix.

Section C: Logistics

17. Time Frame:

Project Start Date	Project Completion Date
09/01/2005	09/01/2007

Dates actively inside the State Marine Refuge.

5/23/2006 to 6/16/2006

Personnel schedule in the State Marine Refuge (describe who will be where and when).

Please refer to question 17 of attached Appendix.

18. Gear and Materials

- [x] Dive equipment
- [ ] Radio Isotopes
- [x] Collecting Equipment
- [x] Chemicals (specify types)

19. Fixed installations and instrumentation.

- [ ] Transect markers
- [ ] Acoustic receivers
- [x] Other (specify)

20. Provide a time line for sample analysis, data analysis, write-up and publication of information.

Please refer to question 20 of attached Appendix.

21. **Vessel Information**:

Vessel Name see attached file for

Vessel Owner vessel information

Captain's Name _____

Vessel Type _____

Length _____

Port of Embarkation _____

IMO Number _____

Flag _____

Chief Scientist or Project Leader _____

Call sign _____

Gross tonnage _____

Last port vessel will have been at prior to this embarkation _____

Total Ballast Water Capacity: Volume _____m3 Total number of tanks on ship _____

Total Fuel Capacity: _____ Total number of fuel tanks on ship _____

Other fuel/chemicals to be carried on board and amounts:

Number of tenders/skiffs aboard and specific type of motors:

Does the vessel have the capability to hold sewage and grey-water? Describe in detail.

Does the vessel have a night-time light protocol for use in the NWHI? Describe in detail (attach additional pages as necessary)

On what workboats (tenders) will personnel, gear and materials be transported within the State Marine Refuge?

Please refer to question 21 of attached Appendix.

How will personnel, gear and materials be transported between ship and shore?

Please refer to question 21 of attached Appendix.

If applicable, how will personnel be transported between islands within any one atoll?

Please refer to question 21 of attached Appendix.

Assessing diversity of microbes associated with healthy and health-compromised corals across the Hawaiian archipelago

Section B: Project Information
8. Project Location
Describe project location (include names, GPS coordinates, attach maps, etc. as appropriate).

Tentative May/June 2006 cruise schedule:

Day	Date	Description / Duration of Stay	Location
1	18-May-06	in transit	Honolulu to Nihoa
2	19-May-06	in transit	Honolulu to Nihoa
3	20-May-06	arrive	Nihoa
4	21-May-06	1 day	Nihoa
5	22-May-06	1 day / overnight transit	Nihoa
6	23-May-06	arrive	French Frigate Shoals
7	24-May-06	1 day	French Frigate Shoals
8	25-May-06	1 day	French Frigate Shoals
9	26-May-06	1 day	French Frigate Shoals
10	27-May-06	1 day / overnight transit	French Frigate Shoals
11	28-May-06	arrive	Gardner Pinnacles
12	29-May-06	1 day	Gardner Pinnacles
13	30-May-06	in transit	Gardner Pinnacles to Johnston Atoll
14	31-May-06	in transit	Gardner Pinnacles to Johnston Atoll
15	1-Jun-06	in transit	Gardner Pinnacles to Johnston Atoll
16	2-Jun-06	arrive	Johnston Atoll
17	3-Jun-06	1 day	Johnston Atoll
18	4-Jun-06	1 day	Johnston Atoll
19	5-Jun-06	1 day	Johnston Atoll
20	6-Jun-06	1 day	Johnston Atoll
21	7-Jun-06	1 day	Johnston Atoll
22	8-Jun-06	in transit	Johnston Atoll to Honolulu
23	9-Jun-06	in transit	Johnston Atoll to Honolulu
24	10-Jun-06	in transit	arrive Honolulu

Organism or objects (List of species, if applicable, add additional sheets if necessary)

Table summarizing tentative dive sites and coral specimen collections for May/June 2006 cruise:

NO. SITES	ATOLL	SITE	LATITUDE Degrees	LATITUDE Minutes	LONGITUDE Degrees	LONGITUDE Minutes	HABITAT	DEPTH (FT.)	COMMON NAME	SCIENTIFIC NAME
1	French Frigate Shoals	TC21	23	50.812 N	-166	19.629 W	fore reef	37	Table coral	*Acropora cytherea*
	French Frigate Shoals	TC21	23	50.812 N	-166	19.629 W	fore reef	37	Lobe coral	*Porites lobata*
2	French Frigate Shoals	R16	23	51.011 N	-166	19.746 W	fore reef	30	Table coral	*Acropora cytherea*
	French Frigate Shoals	R16	23	51.011 N	-166	19.746 W	fore reef	30	Lobe coral	*Porites lobata*
3	French Frigate Shoals	33	23	50.142 N	-166	15.952 W	lagoon	35	Table coral	*Acropora cytherea*
	French Frigate Shoals	33	23	50.142 N	-166	15.952 W	lagoon	35	Lobe coral	*Porites lobata*
4	French Frigate Shoals	23	23	51.943 N	-166	14.382 W	back reef	16	Table coral	*Acropora cytherea*
	French Frigate Shoals	23	23	51.943 N	-166	14.382 W	back reef	16	Lobe coral	*Porites lobata*
5	French Frigate Shoals	H6	23	52.835 N	-166	16.425 W	fore reef	49	Table coral	*Acropora cytherea*
	French Frigate Shoals	H6	23	52.835 N	-166	16.425 W	fore reef	49	Lobe coral	*Porites lobata*
6	French Frigate Shoals	R46	23	46.168 N	-166	15.680 W	coastal	40	Table coral	*Acropora cytherea*
	French Frigate Shoals	R46	23	46.168 N	-166	15.680 W	coastal	40	Lobe coral	*Porites lobata*
7	French Frigate Shoals	12	23	38.278 N	-166	10.779 W	patch reef	59	Table coral	*Acropora cytherea*
	French Frigate Shoals	12	23	38.278 N	-166	10.779 W	patch reef	59	Lobe coral	*Porites lobata*
8	French Frigate Shoals	21	23	50.812 N	-166	19.629 W	patch reef	37	Table coral	*Acropora cytherea*
	French Frigate Shoals	21	23	50.812 N	-166	19.629 W	patch reef	37	Lobe coral	*Porites lobata*
9	French Frigate Shoals	22	23	51.933 N	-166	14.381 W	back reef	40	Table coral	*Acropora cytherea*
	French Frigate Shoals	22	23	51.933 N	-166	14.381 W	back reef	40	Lobe coral	*Porites lobata*
10	French Frigate Shoals	32	23	48.366 N	-166	13.849 W	patch reef	26	Table coral	*Acropora cytherea*
	French Frigate Shoals	32	23	48.366 N	-166	13.849 W	patch reef	26	Lobe coral	*Porites lobata*
11	French Frigate Shoals	34	23	37.682 N	-166	8.122 W	patch reef	30	Table coral	*Acropora cytherea*
	French Frigate Shoals	34	23	37.682 N	-166	8.122 W	patch reef	30	Lobe coral	*Porites lobata*
12	French Frigate Shoals	R29	23	40.711 N	-166	8.791 W	patch reef	31	Table coral	*Acropora cytherea*
	French Frigate Shoals	R29	23	40.711 N	-166	8.791 W	patch reef	31	Lobe coral	*Porites lobata*
13	French Frigate Shoals	R30	23	51.523 N	-166	12.352 W	back reef	5	Table coral	*Acropora cytherea*
	French Frigate Shoals	R30	23	51.523 N	-166	12.352 W	back reef	5	Lobe coral	*Porites lobata*
14	French Frigate Shoals	30	23	50.982 N	-166	17.827 W	patch reef	18	Table coral	*Acropora cytherea*
	French Frigate Shoals	30	23	50.982 N	-166	17.827 W	patch reef	18	Lobe coral	*Porites lobata*
15	Gardner Pinnacles	R3	24	59.812 N	-167	59.929 W	rocky shore	40	Cauliflower coral	*Pocillopora meandrina*
	Gardner Pinnacles	R3	24	59.812 N	-167	59.929 W	rocky shore	40	Rice coral	*Montipora capitata*
	Gardner Pinnacles	R3	24	59.812 N	-167	59.929 W	rocky shore	40	Lobe coral	*Porites lobata*

Additional sites at Nihoa will also be selected. Site selections will be based on accessibility and habitat type.

Table summarizing samples to be shared amongst the Rappé, Gates, Karl, and Toonen labs. The Rappé laboratory will only be using samples from the species shaded in gray.

HIMB coral samples — Species with a sample size of 30 is applicable only if disease and/or bleaching (10 healthy, 10 diseased, 10 bleached) is present for that species, otherwise sample size is reduced to 5.

Coral Species	Site	Number of colonies not to exceed	
Acropora cytherea	FFS	500	* 450 samples are no larger than 2cm^2 (Karl project)
Acropora cytherea	Nihoa	50	
Acropora cytherea	Gardner	50	
Acropora nasuta	FFS	5	
Acropora nasuta	Nihoa	5	
Acropora nasuta	Gardner	5	
Acropora paniculata	FFS	5	
Acropora paniculata	Nihoa	5	
Acropora paniculata	Gardner	5	
Pocillopora damicornis	FFS	5	
Pocillopora damicornis	Nihoa	5	
Pocillopora damicornis	Gardner	5	
Pocillopora meandrina	FFS	50	
Pocillopora meandrina	Nihoa	50	
Pocillopora meandrina	Gardner	50	
Pocillopora eydouxi	FFS	5	
Pocillopora eydouxi	Nihoa	5	
Pocillopora eydouxi	Gardner	5	
Porites lobata	FFS	500	* 450 samples are no larger than 2cm^2 (Karl project)
Porites lobata	Nihoa	50	
Porites lobata	Gardner	50	
Porites brighami	FFS	5	
Porites brighami	Nihoa	5	
Porites brighami	Gardner	5	
Porites lichen	FFS	5	
Porites lichen	Nihoa	5	
Porites lichen	Gardner	5	
Montipora capitata	FFS	500	* 450 samples are no larger than 2cm^2 (Karl project)
Montipora capitata	Nihoa	50	
Montipora capitata	Gardner	50	
Montipora patula	FFS	5	
Montipora patula	Nihoa	5	
Montipora patula	Gardner	5	
Leptastrea bewickensis	FFS	5	
Leptastrea bewickensis	Nihoa	5	
Leptastrea bewickensis	Gardner	5	
Pavona varians	FFS	50	
Pavona varians	Nihoa	50	
Pavona varians	Gardner	50	
Fungia scutaria	FFS	50	
Fungia scutaria	Nihoa	50	
Fungia scutaria	Gardner	50	
Tubastraea coccinea	FFS	50	
Tubastraea coccinea	Nihoa	50	
Tubastraea coccinea	Gardner	50	

* should disease and/or bleaching be encountered, up to 20 healthy, up to 20 diseased and up to 10 bleached fragments will be collected from any

9. Purpose/Need/Scope:
State purpose of proposed research and list hypotheses to be tested:
Purpose:

Microbial pathogens have been identified as the causative agents for a variety of diseases that have devastated coral reefs world wide; however, healthy corals are also known to host diverse and unique communities of microorganisms, including both Bacteria and Archaea. Determining coral disease etiologies is problematic because little is known about the types of microbes found on healthy corals and the functional role that these microbes may play in maintaining host health during disease-free periods. The purpose of this study is to identify the diversity of microbes associated with different species of healthy and health-compromised corals in the Northwestern Hawaiian Islands (NWHI) and to determine the extent to which invasive microbial pathogens are present on NWHI reefs. We will investigate if certain microbes and corals form species-specific associations and whether or not these associations vary on spatial and/or temporal scales. We will also examine if healthy and health-compromised corals of the same species harbor similar or different microbial communities. The ultimate goal of this study is to determine if assessments of microbial community structure can be used as bioindicators of coral health and to predict a coral's susceptibility to disease. This will be the first large scale study of coral associated microbial communities in the NWHI and will be extended to the Main Hawaiian Islands and other locations in the greater Pacific. Microbial communities from these other locales will be compared with those within the NWHI to examine if community structure varies with geographic location and/or anthropogenic input.
Throughout this investigation, the following hypotheses will be tested:

$H_0$1: Coral associated microbial (CAM) community structure (species diversity and relative abundance) does not differ between different coral species.
$H_0$2: CAM community structure does not vary on a micro scale (e.g. within a colony) or on a macro scale (e.g. between different reefs and/or between different islands/atolls).
$H_0$3: CAM community structure does not vary on a seasonal scale.
$H_0$4: Health-compromised (e.g. bleached or diseased) corals do not have different CAM communities than healthy conspecifics.
$H_0$5: Invasive microbial pathogens are not present on NWHI reefs.

Samples for this study have been collected on two previous cruises to the NWHI in May of 2005 and September of 2005. Samples were collected under the auspices and permits of Dr. Greta Aeby from the Hawaii Institute of Marine Biology. The majority of our time during the May 2005 cruise involved aiding Dr. Aeby in the establishment of permanent transects for disease monitoring purposes. When time permitted, minor collections for coral associated microbes were made. During September of 2005, we collected coral samples for our laboratory. Collections made during the May 2006 cruise will enable us to continue our assessment of microbial community structure and invasive microbial pathogens in NWHI corals over spatial and temporal scales.

Table summarizing CAM sample inventory from May 2005 and September 2005 cruises.

DATE	ATOLL	SITE	HABITAT	SPECIES	# COLONIES	# SAMPLES
18-May-05	FFS	TC21	fore reef	*Porites lobata*	2	6
19-May-05	FFS	R16	fore reef	*Porites lobata*	2	6
22-May-05	Maro	TC22	patch reef	*Porites lobata*	1	4
22-May-05	Maro	8	patch reef	*Porites lobata*	3	9
22-May-05	Maro	8	patch reef	*Pocillopora meandrina*	3	9
25-May-05	P&H	TC32	back reef	*Pocillopora meandrina*	3	9
27-May-05	P&H	44	fore reef	*Porites lobata*	3	9
5-Jun-05	FFS	Rapture Reef	fore reef	*Acropora cythaerea*	3	9
17-Sep-05	FFS	33	lagoon	*Porites lobata*	5	15
17-Sep-05	FFS	23	back reef	*Porites lobata*	5	15
18-Sep-05	FFS	H6	fore reef	*Porites lobata*	3	9
18-Sep-05	FFS	R46	coastal	*Porites lobata*	4	12
18-Sep-05	FFS	R46	coastal	*Acropora cytherea*	4	12
19-Sep-05	FFS	12	patch reef	*Acropora cytherea*	11	39
21-Sep-05	Maro	R12	patch reef	*Porites lobata*	7	21
21-Sep-05	Maro	R12	patch reef	*Montipora capitata*	4	8
22-Sep-05	Maro	22	patch reef	*Porites lobata*	7	21
22-Sep-05	Maro	6	patch reef	*Porites lobata*	8	24
22-Sep-05	Maro	6	patch reef	*Montipora capitata*	9	27
24-Sep-05	P&H	31	back reef	*Porites lobata*	10	30
24-Sep-05	P&H	31	back reef	*Montipora capitata*	5	15
25-Sep-05	P&H	R32	back reef	*Porites lobata*	5	15
25-Sep-05	P&H	R31	back reef	*Porites compressa*	7	21
25-Sep-05	P&H	33	fore reef	*Porites lobata*	4	12
25-Sep-05	P&H	R26	fore reef	*Porites lobata*	5	15
26-Sep-05	P&H	26	back reef	*Montipora capitata*	8	36
26-Sep-05	P&H	R39	fore reef	*Porites lobata*	5	15
26-Sep-05	P&H	R44	fore reef	*Porites lobata*	6	21
27-Sep-05	Midway	R3	fore reef	*Porites lobata*	6	18
27-Sep-05	Midway	R7	fore reef	*Porites lobata*	8	24
28-Sep-05	Midway	H21	back reef	*Montipora capitata*	5	3
28-Sep-05	Midway	1	back reef	*Montipora capitata*	6	18
28-Sep-05	Midway	2	fore reef	*Porites lobata*	5	15
29-Sep-05	Kure	2	fore reef	*Porites lobata*	6	18
29-Sep-05	Kure	2	fore reef	*Pocillopora meandrina*	7	21
29-Sep-05	Kure	R33	fore reef	*Porites lobata*	5	15
29-Sep-05	Kure	R33	fore reef	*Pocillopora meandrina*	5	15
29-Sep-05	Kure	R36	back reef	*Pocillopora meandrina*	5	15
30-Sep-05	Kure	14	back reef	*Montipora capitata*	8	24
30-Sep-05	Kure	17	back reef	*Porites lobata*	7	21
30-Sep-05	Kure	18	back reef	*Porites compressa*	5	15
4-Oct-05	Necker	4	rocky shore	*Porites lobata*	10	30
4-Oct-05	Necker	2	rocky shore	*Porites lobata*	5	15
4-Oct-05	Necker	2	rocky shore	*Pocillopora meandrina*	5	15
4-Oct-05	Necker	Shark's Bay	rocky shore	*Porites lobata*	5	15
totals					**245**	**741**

Describe how your proposed project will help provide information or resources to fulfill the National Wildlife Refuge purpose and to reach the Refuge goals and objectives.

Several incidences of coral bleaching and a variety of diseases have been documented in the Northwestern Hawaiian Islands. A mass coral bleaching event was observed in 2002 (Aeby et al. 2003) with another, minor event taking place in 2004. To date, 12 disease states have been recorded in NWHI corals and efforts to elucidate the etiologies of these diseases are ongoing. Disease is a natural part of any ecosystem and fortunately, is not yet present in epizootic proportions in the Hawaiian archipelago based on gross visual inspection. The opportunity to study coral-associated microbial community diversity in an environment far removed from anthropogenic disturbance, such as the NWHI, will allow us to investigate coral health and disease under relatively natural conditions. Comparisons made between studies taking place in the NWHI Marine Sanctuary with those in areas of higher anthropogenic input (e.g. the Main Hawaiian Islands) will enable us to evaluate the impacts of human activities on coral health and disease. This information is of critical importance if we are to effectively address disease prevention in Hawaiian corals by providing appropriate recommendations to resource managers.

Describe context of this research include history of the science for these questions and background of the research.

The current global decline in coral reef health cannot be ignored. The most recent edition of the Current Status of Coral Reefs (Wilkinson 2004) reports that 20% of the world's coral reefs have been destroyed, with severely limited potential for recovery. A predicted 24% of coral reefs world wide are under immediate risk of collapse from anthropogenic disturbances, and 26% are under a longer term threat of collapse. Coral bleaching as a result of global climate change, disease, sedimentation, eutrophication, chemical pollution, coastal development, and over-fishing are listed among the top 10 threats and stressors to coral reefs. The topic of coral disease has been receiving international attention due to an increase in the prevalence of novel pathogenic diseases resulting in the degradation of reefs on a global scale over the past three decades (Lafferty et al. 2004).

There are at least 18 diseases known to affect more than 150 species of Caribbean and Indo-Pacific corals. Etiologies for only 5 out of the 18 known coral diseases have been determined through the fulfillment of Koch's postulates (Sutherland et al. 2004). Known diseases are associated with microbial pathogens including, bacteria, cyanobacteria, fungi and protists and with abiotic stressors including elevated seawater temperature, sedimentation, eutrophication and pollution (Sutherland et al. 2004). Understanding coral disease etiologies is complicated by our lack of understanding of the possible roles that coral-associated microbes play in maintaining coral host health. Healthy corals harbor diverse and abundant microbial communities in their surface mucus layer (Ducklow and Mitchell 1979, Rohwer et al. 2001, 2002, Knowlton and Rohwer 2003, Kellogg 2004, Wegley et al. 2004), in their tissue (Lesser et al. 2004), and possibly within their skeleton (Wafer et al. 1990). The microbes have been proposed to serve several functions including: 1) protection of corals from harmful pathogens through antibiotic production (Geffen and Rosenberg 2005, Harder et al. 2003), 2) providing corals with nutrients (e.g. through nitrogen fixation), and 3) providing an additional food source (Rohwer et al. 2002). Species-specific associations appear to be present between some bacteria and coral species (Rohwer et al. 2001, 2002); however, whether or not these two organisms engage in a mutualistic symbiosis is yet to be determined. To date, few studies have examined the functional relationships between microbes and the coral host organism and its symbiotic dinoflagellates.

Before we can begin to address the multitude of diseases afflicting corals world wide, it is absolutely essential to first identify the microbial communities associated with healthy corals.

The current trend of mediating coral disease is akin to diagnosing a sick human patient without any knowledge of the biological statistics of the patient when he or she is in a healthy state. Recent studies have shown that microbial community structure on healthy corals may differ between species and that certain phylotypes of bacteria are present on all individuals of a particular species (Rohwer et al. 2001, 2002). Depending on the composition of microbes and their interaction with the host coral, certain coral-microbe associations may respond differently to environmental disturbances, rendering the host coral to be more or less susceptible to disease. A study by Klaus et al. (2005) showed that the most abundant bacterial taxa associated with healthy *Diploria strigosa* exhibited increased dominance at sites adjacent to a sewage outlet compared to conspecifics at non-impacted sites. Conversely, healthy *Montastraea annularis* colonies located along the same gradient of anthropogenic input did not exhibit differences in microbial community structure. Environmental disturbances that alter the composition of the "healthy state" microbial community, which has been proposed to serve a protective role, may compromise the coral's resistance to invasion by microbial pathogens. Pantos et al. (2003) observed significant differences amongst microbial communities associated with disease lesions in *Montastraea annularis*, apparently healthy coral tissue distant from the disease lesions, and tissue from healthy colonies of this species. It is also possible that environmental stressors offset the dynamics of the coral-microbe association, allowing certain microbes present during the healthy state to opportunistically dominate over others when the coral is under stress, leading to disease. Investigations by Friaz-Lopez et al. (2004), to determine the composition of black band disease bacterial mats, revealed that 6 out of 8 bacterial phylotypes consistently abundant in the mats were also consistently detected in healthy corals, with 4 of the 6 bacteria being more abundant in black band disease mats than in healthy corals. Findings from the aforementioned studies suggest that examining the structure of microbial communities associated with corals may be useful for predicting a coral species' susceptibility to disease as well as for detecting physiological stress before any physical signs of disease are present.

Explain the need for this research and how it will help to enhance survival or recovery of refuge wildlife and habitats.

Gaining a solid understanding of the diseases that affect Hawaii's corals is essential if we are to effectively prevent them from causing widespread harm to Hawaii's coral reef ecosystems. The methods employed in this study may enable us to predict a coral's susceptibility to disease following an environmental disturbance. If we are able to determine which coral species are the most sensitive to particular environmental disturbances, we can potentially mediate these disturbances to enhance the survival or recovery of this species. Most importantly, we believe that our study methods will enable us to detect physiological stress in a coral before the onset of disease occurs. By identifying the early warning signs of disease, we can potentially prevent disease from spreading to epizootic proportions by developing methods to effectively treat or quarantine diseased corals. Finally, this study should enable us to identify invasive microbial pathogens, determine their source, and begin to develop methods that will prevent them from entering the pristine coral reef ecosystems of the NWHI.

Describe how your proposed project can help to better manage the National Wildlife Refuge or global communities.

Developing methods that enable scientists to predict a coral's health state and susceptibility to disease will, in turn, foster the development of methods that will enable resource managers to better protect corals from disease. Disease prevention management may include establishing protected areas for more sensitive, disease prone coral species, as well as for coral species that are more robust and resilient to disease, reducing human impacts that may

increase a coral's susceptibility to disease, prohibiting the introduction of any foreign materials or pollutants that may alter coral-associated microbial communities, and other measures that will help to prevent the spread of disease.

10. Procedures (include equipment/materials)

Coral species that are common and ecologically important in the Northwestern Hawaiian Islands, and throughout the Pacific, have been selected for microbial community analysis. These inlcude: *Porites lobata*, *Porites compressa*, *Pocillopora meandrina*, *Montipora capitata* and *Acropora cytherea*. Using SCUBA and underwater tools, three non-lethal sub-samples are taken from each coral colony, with at least 5 colonies sampled per species per site (we expect to collect approximately the same number of samples retrieved during the September 2005 cruise). A sub-sample consists of a 6-mm diameter, 6-mm deep core (similar in size to a fish bite) that includes the coral tissue, overlying mucus layer, and underlying skeleton. After collection in the field, core samples are killed by freezing on board the ship. Our sampling strategy focuses on collecting from healthy coral colonies, with diseased and/or bleached colonies being sampled when encountered (During the September 2006 cruise, we would like to collect from healthy and health-compromised coral colonies at French Frigate Shoals, where Dr. Greta Aeby has documented the presence of Acropora White Syndrome disease in *Acropora cytherea* corals). Samples are collected from various habitats (e.g. fore reef, back reef, lagoon) at each atoll/island. Seawater samples are taken adjacent to sampled coral heads for comparison of CAM community composition between corals and the surrounding seawater environment. Other environmental variables including temperature, salinity, nutrients, and cholorphyll a will be measured in the water column. CAM samples are processed and stored in the -37°C freezer aboard the research vessel. All samples are then transported to the Hawaii Institute of Marine Biology (HIMB) on Coconut Island for further processing and molecular analysis in the laboratory of Dr. Michael Rappé. Samples collected from the cruise will be integrated into an on-going time-series study of seasonal changes in CAM diversity in the MHI and NWHI.

In the laboratory, CAM communities associated with each coral species are assessed through molecular analyses of microbial 16S rRNA genes. Molecular tools enable us to identify microorganisms that are not detected using traditional culturing methods. Techniques for CAM DNA extraction and amplification have been optimized for each target coral species. Polymerase chain reaction-amplified CAM DNA is used in a community fingerprinting technique known as terminal restriction fragment length polymorphism (TRFLP) to assess the diversity and relative abundance of microbial species associated with each individual coral sample. CAM PCR products will also be used in cloning and sequencing to identify microbes down to species or phylotype. In addition, currently available nucleic acid sequence and community fingerprinting data is being compiled into a database for the rapid identification of microbial taxa based on phylogenetically-informative and discriminative signatures.

12. Literature cited

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13. What types of insurance do you have in place?

NOAA Ship HI`IALAKAI is a U.S. Government-owned and –operated research vessel and is self-insured by the U.S. Government.

14. What certifications/inspections do you have scheduled for your vessel?

Rat Free (scheduled with U.S. Dept. of Health and Human Services for April 2006)

Hull Inspection (scheduled with Hawaii Institute of Marine Biology biologists (normally Scott Godwin) prior to projects working in the Northwestern Hawaiian Islands (NWHI)) to ensure no nuisance algae or other fouling species are transported to the NWHI.

Ballast water information is transmitted to USCG as required by CFR Title 33, Vol. 2, Parts 151.1500 to 199; IMO Resolution A.868(20); and USCG COMDTPUB P16700.4

16. Project's relationship to other research projects within the NWHI State Marine Refuge, National Wildlife Refuge, NWHI Coral Reef Ecosystem Reserve, or elsewhere.

Project Title: Investigation of bleached and diseased corals in the Northwestern Hawaiian Islands
Principal Investigator: Greta S. Aeby, Hawaii Institute of Marine Biology
This project will compliment Dr. Aeby's work on characterizing white syndrome disease in Acropora cytherea at French Frigate Shoals.

Project Title: Extrinsic and intrinsic factors affecting the resilience of corals to climate, change, and their use in designing marine reserves
Principal Investigator: Dr. Charles Birkeland, Assistant Leader, Hawaii Cooperative Fishery Research Unit and Adjunct Associate Professor, Zoology Department, University of Hawaii at Manoa
Microbial diversity will be assessed in corals found in Ofu lagoon, American Samoa. Like the NWHI, Ofu is relatively removed from anthropogenic disturbances. Microbial

communities identified in Ofu coral species will be compared to those found on coral species in the NWHI.

17. Personnel schedule in the State Marine Refuge (describe who will be where and when).

Jennifer L. Salerno will participate in the May/June cruise. She is a Ph.D. student in Zoology/EECB at the University of Hawaii in Manoa and an AAUS-certified scientific diver. Jennifer has participated in two previous cruises to the NWHI National Wildlife Refuge where she has participated in coral disease assesment, establishing permanent monitoring transects, and coral specimen collection. Anderson B. Mayfield will also participate in specimen collections during the May/June cruise. He is a Ph.D. student in Zoology at the University of Hawaii in Manoa and an AAUS-certified scientific diver. Both participants have signficant field work and diving experience in remote temperate and tropical marine ecosystems.

18. Gear and Materials

Chemicals:
95% Ethanol (MSDS sheets attached)
10% paraformaldehyde (MSDS sheets attached)

19. Fixed installations and instrumentation.

We would like to mark approximately 10 colonies of *Acropora cytherea* 10 colonies of *Porites lobata* using three plastic zip ties per colony. Marking the colonies will enable divers to relocate and sample individual colonies in September and October as part of a time-series seasonal study of the coral associated microbial communities. Colonies will be marked in areas where permanent transects have been set up by Dr. Greta Aeby. This will allow for easy relocation of the colonies. Plastic markers will be removed immediately after the study is completed in October.

20. Provide a time line for sample analysis, data analysis, write-up and publication of information.

June 2006 - August 2006: Molecular analyses of coral associated microbial communities in the laboratory
August 2006 - January 2007: Data analysis and write-up of results for publication in a peer-reviewed scientific journal

21. Vessel Information

Vessel Name – NOAA Ship HI`IALAKAI
IMO Number – 8835619
Vessel Owner – U.S. Dept. of Commerce, National Oceanic and Atmospheric Administration (NOAA)
Flag – USA
Captain's Name – CDR Scott Kuester, NOAA
Chief Scientist or Project Leader – Randall Kosaki, Ph.D., NOAA
Vessel Type – Oceanographic Research
Call Sign – WTEY
Length – 224 feet
Gross Tonnage – 1,914
Port of Embarkation – Honolulu

Last port vessel will have been at prior to this embarkation – Pago Pago, Amer. Samoa
Total Ballast Water Capacity:
Volume – 487 m3 (128,834 U.S. gal.)
Total number of ballast tanks on ship – 10
Total Fuel Capacity:
228,642 U.S. gal. at 98% capacity
Total number of fuel tanks on ship – 15
Other fuel/chemicals to be carried on board and amounts: gasoline – as much as 700 U.S. gal.; lube oil – as much as 10,442 U.S. gal.; numerous other industrial and household chemicals used to operate a 224-foot research vessel
Number of tenders/skiffs aboard and specific type of motors:
Ship's own tenders - 1 each 10 m AMBAR Marine jet boat with Yanmar 370-hp, Diesel inboard engine
1 each 8 m AMBAR Marine jet boat with Yanmar 315-hp, Diesel inboard engine
2 each 17.5 ft Zodiac inflatable boats, each with one Honda 50-hp, 4-stroke, outboard gasoline engine
1 each 19 ft AMBAR Marine rescue boat with Honda 115-hp, 4-stroke, outboard gasoline engine

Program-provided tenders – 19' Boston Whaler with 135 hp Honda four-stroke outboard

Does the vessel have the capability to hold sewage and grey-water? Describe in detail. The ship has a 4,000 U.S. gal Collection Holding Tank for sewage and grey water. In those waters where effluent may NOT be discharged, sewage and grey water are held in this tank until the ship is in waters where sewage and grey water may be discharged. The ship has a U.S. Coast Guard-approved Marine Sanitation Device (Omnipure model MSD 12 MC) which is used to treat sewage and grey water in those waters where effluent may be discharged.

Does the vessel have a night-time light protocol for use in the NWHI? Describe in detail. Navigation lights are on 24-hours/day. Work lights are put on at night only when conducting CTD operations. Weather decks are not illuminated at night.

On what workboats (tenders) will personnel, gear and materials be transported within the State Marine Refuge? - Personnel, gear and materials may be transported within the State Marine Refuge by the ship or any of the 5 ship's small boats listed above or by the program-provided small boat listed above.

How will personnel, gear and materials be transported between ship and shore? – Personnel, gear and materials may be transported between ship and shore by any of the 5 ship's small boats listed above or by the program-provided small boat listed above.

If applicable, how will personnel be transported between islands within any one atoll? - Personnel may be transported between islands within any one atoll by any of the 5 ship's small boats listed above or by the program-provided small boat listed above.

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Education

Ph.D., Genetics Program and Department of Microbiology, Oregon State University. Academic Advisor: Dr. Stephen Giovannoni. Dissertation: "Diversity of bacterioplankton and plastid SSU rRNA genes from the eastern and western continental shelves of the United States"	1997
B.S., Biology and University Honors Program, Washington State University.	1991

Professional Experience

Assistant Research Professor, Hawaii Institute of Marine Biology, School of Ocean and Earth Science and Technology, University of Hawaii at Manoa.	7/03-present
Postdoctoral Research Associate, High Throughput Culturing Facility, Department of Microbiology, Oregon State University. Advisor: Dr. Stephen Giovannoni.	3/99-6/03
Postdoctoral Research Associate, Anaerobic Microbiology Group, College of Veterinary Medicine, Oregon State University. Advisor: Dr. A. Morrie Craig.	3/99-7/00
Postdoctoral Research Associate, Phytoplankton Group, Centre National de la Recherche Scientifique, Station Biologique de Roscoff, France. Advisor: Dr. Daniel Vaulot.	6/97-3/99
Graduate Research Assistant, Department of Microbiology and Genetics Program, Oregon State University. Advisor: Dr. Stephen Giovannoni.	8/93-5/97
Undergraduate Research Assistant, Department of Zoology, Washington State University.	1989-1991
Undergraduate Research Assistant, Department of Veterinary Medicine, Washington State University.	1987-1988

Teaching Experience

Co-Instructor, Coral Molecular Biology Techniques Workshop, Hawaii Institute of Marine Biology, University of Hawaii. Responsibilities: lecturing, organize and lead laboratory and field exercises.	Summers, 2003-present
Guest Lecturer, Astrobiology Winter School, University of Hawaii NASA Astrobiology Institute,. Responsibilities: lecturing, lead discussion groups.	January 2005
Co-Instructor, Marine Microplankton Ecology (OCN 626), Department of Oceanography, University of Hawaii. Responsibilities: lecturing, organize and lead discussion group.	Falls, 2004-present
Supervisory and Training, University of Hawaii and Oregon State University.	Ongoing

Responsibilities: train graduate and undergraduate students in laboratory techniques, analysis of nucleic acid sequence data, notebook keeping, communications skills, scientific writing, and science ethics; aid in supervision of research activity in the laboratory.

Co-Instructor, Microbial Diversity (MB 420/520), Department of Microbiology, Oregon State University. Responsibilities: course development including syllabus, lecture notes, and exams; organized and lead weekly discussion groups for graduate students; lecturing. Spring 2001-3

Co-Instructor, Genomics and Cellular Evolution (MB 668), Department of Microbiology, Oregon State University. Responsibilities: course development including syllabus, lecture notes, and exams; lecturing; organize, lecture, and guide students through weekly computer laboratory sessions; individual assistance with bioinformatics course projects. Winter 2001-2

National and International Professional Activities and Service

Professional affiliations: American Society for Microbiology, American Society of Limnology and Oceanography, International Society for Microbial Ecology, American Association for the Advancement of Science.

Peer reviewer: *Science, Microbiology and Molecular Biology Reviews, Applied and Environmental Microbiology, Limnology and Oceanography, Limnology and Oceanography Methods, FEMS Microbiology Ecology, Aquatic Microbial Ecology, Proceedings of the Royal Society, London: Biological Sciences, BioTechniques, Trends in Microbiology, Journal of Geophysical Research-Biogeosciences*, NSF, NASA, NERC, NOAA.

Panelist: NSF Biogeosciences (2003), NASA Astrobiology (2003).

Technical Advisory Committee, Oregon/Hawaii Sea Grant Marine Biotech Outreach Project, 2004-present.

Co-Chair, Topical Session 54, *Advances in Microbial Ecology: New Culturing Techniques and Culture-independent Approaches*, 2005 ASLO Aquatic Sciences Meeting, 20-25 February 2005, Salt Lake City, Utah.

Invited participant: US National Oceanographic Partnership Program, Ocean Ecogenomics Meeting, 7-8 March 2005, Washington DC.

Invited participant: American Academy of Microbiology, Marine Microbial Diversity: The Key to Earth's Habitability Colloquium, 8-10 April 2005, San Francisco, California.

Invited participant: Alfred P. Sloan Foundation, International Census of Marine Microbes Open Oceans and Coastal Systems Workshop, 10-11 May 2005, Honolulu, Hawaii.

University and Departmental Professional Activities and Service

Graduate Faculty Status in University of Hawaii Manoa Departments of Oceanography and Microbiology.

Current graduate student committees (*primary advisor): Amy Apprill* (Ph.D., UH Oceanography), Marina Brandon* (M.S., UH Oceanography), Darin Hayakawa* (Ph.D., UH Microbiology), Jennifer Salerno* (Ph.D., UH Zoology), Sara Yeo* (M.S., UH Oceanography), Tracy Campbell* (M.S., UH Oceanography), Jillian Ward (Ph.D., UH Oceanography), Olivia Mason (Ph.D., OSU Oceanography).

Past graduate student committees (*primary or co-advisor): Marcie Workman (M.S., UH Oceanography).

Undergraduate student research supervision: Lei Young (Oregon State University, Dept. of Microbiology, 2002-03), Friederike Baumann (Anhalt University, Germany, 2003-04), Cornelia Schmidt (Anhalt University, Germany, 2003-04), Sara Yeo (Hawaii Pacific University, 2004-05), Tracy Campbell (University Wisconsin Stout, 2005), Elizabeth Hambleton (Williams College, 2005), Hollie Kerr (California State University, 2005).

Faculty Judge, Student Travel for Achievement in Research (STAR) Awards, UH Manoa Department of Oceanography, 2005.

Leonida Scholarship Selection Committee, UH Manoa SOEST, 2004.

Search Committee, NWHICRER Grant Administrator, HIMB, 2005.

Search Committee, Evolutionary/Conservation Geneticist Faculty Position, HIMB, 2004.

Search Committee, Marine Sciences Faculty Position, HIMB, 2005.

Publications

1. Giovannoni, S. J., L. Bibbs, J.-C. Cho, M. D. Staples, R. Desiderio, K. L. Vergin, **M. S. Rappé**, S. Laney, D. F. Barofsky, and E. Mathur (2005) Proteorhodopsin phototrophy in the ubiquitous marine bacterium SAR11. Nature 438:82-85.
2. Giovannoni, S. J., H. J. Tripp, S. Givan, M. Podar, K. L. Vergin, D. Baptista, L. Bibbs, J. Eads, T. H. Richardson, M. Noordewier, **M. S. Rappé**, J. M. Short, J. C. Carrington and E. J. Mathur (2005) Genome streamlining in a cosmopolitan oceanic bacterium. Science 309:1242-1245.
3. Morris, R. M., J.-C. Cho, K. L. Vergin, **M. S. Rappé**, C. A. Carlson, and S. J. Giovannoni (2005) Temporal and spatial response of bacterioplankton lineages to annual convective overturn at BATS. Limnology and Oceanography 50:1687-1696.
4. Lodge-Ivey, S. L., **M. S. Rappé**, W. H. Johnston, R.E. Bohlken, and A. M. Craig (2005) Molecular analysis of a consortium of ruminal microbes that detoxify pyrrolizidine alkaloids. Canadian Journal of Microbiology 51:455-465.
5. Morris, R. M., **M. S. Rappé**, S. A. Connon, E. Urbach, and S. J. Giovannoni (2004) Prevalence of the *Chloroflexi*-related SAR202 bacterioplankton cluster throughout the mesopelagic zone and deep ocean. Applied and Environmental Microbiology 70:2836-2842.
6. **Rappé, M. S.**, and S. J. Giovannoni (2003) The uncultured microbial majority. Annual Review of Microbiology 57:369-394.
7. Cowen, J. P, S. J. Giovannoni, F. Kenig, H. P. Johnson, D. Butterfield, **M. S. Rappé**, M. Hutnak, and P. Lam (2003) Fluids from aging ocean crust that support microbial life. Science 299:120-123.
8. Zengler, K., G. Toledo, **M. Rappé**, J. Elkins, E. Mathur, J. Short, and M. Keller (2002) Cultivating the uncultured. Proceedings of the National Academy of Sciences, USA 99:15681-15686.
9. Morris, R. M., **M. S. Rappé**, S. A. Connon, K. L. Vergin, W. A. Siebold, C. Carlson, and S. J. Giovannoni (2002) SAR11 clade dominates ocean surface bacterioplankton communities. Nature 420:806-810.

10. **Rappé, M. S.**, S. A. Connon, K. L. Vergin, and S. J. Giovannoni (2002) Cultivation of the ubiquitous SAR11 marine bacterioplankton clade. Nature 418:630-633.

11. Hold, G. L., E. A. Smith, **M. S. Rappé**, E. W. Maas, E. R. B. Moore, C. Stroempl, J. R. Stephen, J. I. Prosser, T. H. Birkbeck, and S. Gallacher (2001) Characterization of bacterial communities associated with toxic and non-toxic dinoflagellates: *Alexandrium* spp. and *Scrippsiella trochoidea*. FEMS Microbiology Ecology 37:161-173.

12. Vergin, K. L., **M. S. Rappé**, and S. J. Giovannoni (2001) Streamlined method to analyze 16S rRNA gene clone libraries. BioTechniques 30:938-944.

13. Brinkmeyer, R., **M. Rappé**, S. Gallacher, and L. Medlin (2000) Development of clade (*Roseobacter* and *Alteromonas*) and taxon-specific oligonucleotide probes to study bacterial/toxic algal interactions in toxic dinoflagellate cultures. European Journal of Phycology 35:315-329.

14. **Rappé, M. S.**, K. Vergin, and S. J. Giovannoni (2000) Phylogenetic comparisons of a coastal bacterioplankton community with its counterparts in open ocean and freshwater systems. FEMS Microbiology Ecology 33:219-232.

15. **Rappé, M. S.**, D. Gordon, K. Vergin, and S. J. Giovannoni (1999) Phylogeny of Actinobacteria small subunit (SSU) rRNA gene clones recovered from marine bacterioplankton. Systematic and Applied Microbiology 22:106-112.

16. Suzuki, M. T., **M. S. Rappé**, and S. J. Giovannoni (1998) Kinetic bias in estimates of coastal picoplankton community structure obtained by measurements of small-subunit rRNA gene PCR amplicon length heterogeneity. Applied and Environmental Microbiology 64:4522-4529.

17. **Rappé, M. S.**, M. T. Suzuki, K. Vergin, and S. J. Giovannoni (1998) Phylogenetic diversity of ultraphytoplankton plastid SSU rRNA genes recovered in environmental nucleic acid samples from the Pacific and Atlantic coasts of the United States. Applied and Environmental Microbiology 64:294-303.

18. **Rappé, M. S.**, P. F. Kemp, and S. J. Giovannoni (1997) Phylogenetic diversity of marine coastal picoplankton 16S rRNA genes cloned from the continental shelf off Cape Hatteras, North Carolina. Limnology and Oceanography 42:811-826.

19. Suzuki, M. T., **M. S. Rappé**, Z. W. Haimberger, H. Winfield, N. Adair, J. Ströbel, and S. J. Giovannoni (1997) Bacterial diversity among SSU rDNA gene clones and cellular isolates from the same seawater sample. Applied and Environmental Microbiology 63:983-989.

20. Field, K. G., D. Gordon, **M. S. Rappé**, E. Urbach, T. Wright, and S. J. Giovannoni (1997) Diversity and depth-specific distribution of SAR11 cluster rRNA genes from marine planktonic bacteria. Applied and Environmental Microbiology 63:63-70.

21. Giovannoni, S. J., **M. S. Rappé**, K. Vergin, and N. Adair (1996) 16S rRNA genes reveal stratified open ocean bacterioplankton populations related to the Green Non-Sulfur bacteria. Proceedings of the National Academy of Sciences, USA 93:7979-7984.

22. **Rappé, M. S.**, P. F. Kemp, and S. J. Giovannoni (1995) Abundant chromophyte plastid 16S ribosomal RNA genes found in a clone library from Atlantic Ocean seawater. Journal of Phycology 31:979-988.

Book Chapters

1. Giovannoni, S. J., and **M. S. Rappé** (2000) Evolution, diversity and molecular ecology of marine prokaryotes. *In* D. L. Kirchman [ed.], Microbial Ecology of the Oceans. Wiley and Sons, pp. 47-84.
2. Giovannoni, S., and **M. Rappé** (1999) Microbial diversity: it's a new world. The NEB Transcript 10:1-4.
3. Giovannoni, S. J., **M. S. Rappé**, D. Gordon, E. Urbach, M. Suzuki, and K. G. Field. Ribosomal RNA and the evolution of bacterial diversity (1996) *In* Roberts, D. McL., P. Sharp, G. Alderson, and M. Collins [eds], Evolution of microbial life. Society for General Microbiology Symposium 54. Cambridge University Press, pp. 63-85.

Manuscripts in Preparation

1. Brandon, M. L., J. W. Becker and **Rappé, M. S.** (2006) Cultivating micro-organisms from dilute aquatic environments: melding traditional methodology with new cultivation techniques and molecular methods. For submission to Manual of Environmental Microbiology, 3rd Edition. Invited chapter.
2. Brandon, M. L., and **M. S. Rappé** (2006) Harnessing uncultivated microbes. For submission to Current Opinion in Microbiology. Invited review.
3. Brandon, M. L., J. W. Becker and **M. S. Rappé** (2006) Isolation of aquatic microorganisms via high throughput cultivation methods. For submission to Molecular Microbial Ecology Manual, 3rd Edition. Invited chapter.
4. Salerno, J., A. Apprill, E. Hambleton, and **M. S. Rappé** (2006) Optimization of nucleic acid extraction and PCR methods for coral-associated microorganisms. For submission to Molecular Ecology.
5. Glazer, B. T., J. P. Cowen, D. Copson, D. Harris, J. Jolly, D. B. Nuzzio, J. W. Becker and **M. S. Rappé** (2006) A seafloor observatory package for in situ geochemical measurements concurrent to in situ filtration. For submission to Deep Sea Research.

Presentations and Abstracts

1. Becker, J. W., and **M. S. Rappé**. Mesoscale to microscale: molecular microbial ecology within cyclonic eddies in the lee of the Hawaiian Islands. Poster to be presented at the 2006 Ocean Sciences Meeting, Honolulu, Hawaii, USA, 20-24 February 2006.
2. Hayakawa, D. H, and **M. S. Rappé**. Isolation and molecular ecology of novel Beta Proteobacteria from sub-tropical coastal seawater. Talk to be presented at the 2006 Ocean Sciences Meeting, Honolulu, Hawaii, USA, 20-24 February 2006.
3. Brandon, M. L., and **M. S. Rappé**. High-throughput isolation of marine bacterioplankton from Kane`ohe Bay, Hawaii. Talk to be presented at the 2006 Ocean Sciences Meeting, Honolulu, Hawaii, USA, 20-24 February 2006.
4. Salerno, J. L., A. Apprill, E. Hambleton, and **M. S. Rappé.** Assessing the composition of microbial communities associated with colonies of diverse coral species in the main and Northwestern Hawaiian Islands. Talk to be presented at the 2006 Ocean Sciences Meeting, Honolulu, Hawaii, USA, 20-24 February 2006.
5. Glazer, B. T., J. P. Cowen, D. Copson, D. Harris, J. Jolly, D. B. Nuzzio, J. W. Becker and **M. S. Rappé.** A seafloor instrument package for in situ geochemical measurements

concurrent to in situ filtration. Poster to be presented at the 2006 Ocean Sciences Meeting, Honolulu, Hawaii, USA, 20-24 February 2006.

6. **Rappé, M. S.** Expanding the traditional cultivation-based repertoire to isolate bacterioplankton for new model biosystems in microbial oceanography. Invited tutorial lecture presented at the 2005 ASLO Aquatic Sciences Meeting, Salt Lake City, Utah, USA, 20-25 February 2005.
7. **Rappé, M. S.** Melding cultivation-based and molecular ecological approaches to isolate and study new model biosystems in microbial oceanography. Invited seminar presented to the Department of Microbiology, Cornell University, Ithaca, New York, 27 January 2005.
8. **Rappé, M. S.** An introduction to the ARB sequence analysis package. Invited workshop presented at Cornell University, Ithaca, New York, 26 January 2005.
9. Zengler, K., G. Clark, I. Haller, G. Toledo, M. Walcher, **M. Rappé**, G. Woodnut, J. M. Short, M. Keller. Accessing microbial diversity by high throughput cultivation. Talk presented at the 10th International Symposium on Microbial Ecology, Cancun, Mexico, 22-27 August 2004.
10. **Rappé, M. S.** Marine microbial ecology: Linking global processes to microbial community structure and genomes. Invited talk presented at the Marine Biotechnology Outreach and Education Project Curriculum Development Workshop, Oregon and Hawaii SeaGrant, Hawaii Institute of Marine Biology, Kaneohe, Hawaii, USA, 28 July 2004.
11. Morris, R. M., **M. S. Rappé**, J. C. Cho, K. Vergin, C. A. Carlson, and S. J. Giovannoni. Seasonal shifts in bacterioplankton community structure at BATS. Poster presented at the 104th General Meeting of the American Society for Microbiology, New Orleans, Louisiana, USA, 23-27 May 2004.
12. **Rappé, M. S.** Using over 10 years of rRNA data to guide a new generation of cultivation- and genomics-based research in marine microbial ecology. Invited talk presented at the Department of Zoology, University of Hawaii at Manoa, Honolulu, Hawaii, USA, 30 April 2004.
13. Morris, R. M., **M. S. Rappé**, K. L. Vergin, J. Oliver, R. Parsons, C. A. Carlson, and S. J. Giovannoni. In situ and experimental dynamics of group specific bacterioplankton: recent highlights from an oceanic microbial observatory. Poster presented at the Second Microbial Observatories Principal Investigators' Workshop, Washington DC, USA, 14-16 September 2003.
14. Morris, R. M., T. D. Wright, E. Urbach, **M. S. Rappé**, K. L. Vergin, S. A. Connon, and S. J. Giovannoni. Direct cell counts reveal mesopelagic distribution of the Green Non-Sulfur (GNS) related SAR202 bacterioplankton group. Talk presented at the 2003 ASLO Aquatic Sciences Meeting, Salt Lake City, Utah, USA, 9-14 February 2003.
15. **Rappé, M. S.** The SAR11 marine bacterioplankton group: cultivation of one of the most abundant organisms on Earth. Invited talk presented at the University of Hawaii, School of Ocean and Earth Science and Technology, Honolulu, Hawaii, USA, 24 June 2002.
16. **Rappé, M. S.,** and S. J. Giovannoni. The major marine bacterioplankton rRNA groups: diversity, distribution, dynamics and cultivation. Invited talk presented at the 2002 ASLO Summer Meeting, Victoria, B.C., 10-14 June 2002.
17. **Rappé, M. S.,** R. M. Morris, K. L. Vergin, and S. J. Giovannoni. *Pelagibacter ubique* gen. nov., sp. nov., characterization of the first isolates from the SAR11 marine bacterioplankton clade from the Pacific Ocean. Poster presented at the 102nd General

Meeting of the American Society for Microbiology, Salt Lake City, Utah, USA, 17-21 May 2002.

18. Alexander, C. A., **M. S. Rappé**, K. L. Vergin, and S. J. Giovannoni. Identification of marine bacteria associated with phytoplankton blooms in Oregon coastal waters. Poster presented at the 102nd General Meeting of the American Society for Microbiology, Salt Lake City, Utah, USA, 17-21 May 2002.
19. **Rappé, M. S.,** S. A. Connon, K. L. Vergin, and S. J. Giovannoni. Cultivation of the ubiquitous marine bacterium SAR11. Talk presented at the 2002 Ocean Sciences Meeting, Honolulu, Hawaii, USA, 11-15 February 2002.
20. Morris, R. M., S. A. Connon, **M. S. Rappé**, K. L. Vergin, W. A. Siebold, C. Carlson, and S. J. Giovannoni. In-situ abundance of the SAR11 bacterioplankton clade in the North Atlantic Ocean. Talk presented at the 2002 Ocean Sciences Meeting, Honolulu, Hawaii, USA, 11-15 February 2002.
21. Giovannoni, S., and **M. S. Rappé**. Patterns in the distributions of the marine bacterial groups in the oceans. Talk presented at the 9th International Symposium on Microbial Ecology, Amsterdam, The Netherlands, 26-31 August 2001.
22. Smith, E. A., J. Macrae, F. Grant, S. Gallacher, G. L. Hold, and **M. S. Rappé**. Characterization and phylogenetic diversity of bacteria associated with toxic and non-toxic algae. Talk presented at the 7th European Marine Microbiology Symposium, Noordwijkerhout, The Netherlands, 17-22 September 2000.
23. Brinkmeyer, R., **M. Rappé**, K. Töbe, D. Vaulot, S. Gallacher, and L. K. Medlin. Development of clade (*Roseobacter* and *Alteromonas*) and species-specific oligonucleotide probes to study bacterial/algal interactions and their role in Harmful Algal Bloom ecology. Poster presented at the 2nd European Phycological Conference, Montecatini Terme, Italy, 20-26 September 1999.
24. Smith, E. A., G. L. Hold, **M. Rappé**, C. M. J. Ferguson, F. Milne, and S. Gallacher. Enumeration and phylogenetic analysis of bacteria associated with toxic and non-toxic dinoflagellates: *Alexandrium* spp. and *Scrippsiella trochoidea*. Talk presented at the Aquatic Associations of the UK Conference "Microbial Aquatic Symbiosis: From Phylogeny to Biotechnology," Oban, Scotland, 1-3 September 1999.
25. Giovannoni, S. J., **M. Rappé**, and B. Lanoil. What we have learned from a decade of studying picoplankton diversity using molecular techniques, and the application of this knowledge to the identification of single cells. Talk presented at the 1999 ASLO Aquatic Sciences Meeting, Santa Fe, New Mexico, USA, 1-5 February 1999.
26. **Rappé, M. S.**, G. Hold, S. Gallacher, R. Brinkmeyer, M. Lange, L. K. Medlin, and D. Vaulot. Diversity and population dynamics of bacteria in co-culture with the marine dinoflagellates *Scrippsiella trochoidea* and *Alexandrium tamarense*. Poster presented at the 98th General Meeting of the American Society for Microbiology, Atlanta, Georgia, USA, 17-21 May 1998.
27. **Rappé, M. S.**, K. Vergin, and S. J. Giovannoni. Diversity of plastid-derived 16S rRNA genes cloned from bulk environmental DNA. Poster presented at the 1st European Phycological Congress, Cologne, Germany, 11-18 August 1996.
28. Giovannoni, S. J., and **M. S. Rappé**. Molecular ecological approaches for assessing phytoplankton diversity and population dynamics. Talk presented at the 1st European Phycological Congress, Cologne, Germany, 11-18 August 1996.

29. Suzuki, M. T., Z. W. Haimberger, **M. S. Rappé**, H. Winfield, J. Ströbel, and S.J. Giovannoni. A comparison between the 16S rRNA of marine bacterioplankton strains isolated from the Oregon coast to those cloned from bulk DNA from the same water sample. Talk presented at the 7th International Symposium on Microbial Ecology, Sao Paulo, Brazil, 27 August - 1 September 1995.

30. Giovannoni, S. J., M. T. Suzuki, and **M. S. Rappé**. Bacterioplankton diversity: uncultured or unculturable? Talk presented at the 7th International Symposium on Microbial Ecology, Sao Paulo, Brazil, 27 August - 1 September 1995.

31. **Rappé, M. S.**, and S. J. Giovannoni. Analysis of marine coastal picoplankton 16S rRNA genes cloned from the continental shelf at Cape Hatteras. Poster presented at the 95th General Meeting of the American Society for Microbiology, Washington DC, USA, 21-25 May 1995.

32. **Rappé, M. S.**, P. F. Kemp, and S. J. Giovannoni. Abundant chromophyte plastid 16S ribosomal RNA genes found in a clone library from Atlantic Ocean seawater. Poster presented at the 9th Northwest Algal Symposium, Anacortes, Washington, USA, 31 March - 2 April 1995.

33. **Rappé, M. S.**, P. F. Kemp, and S. J. Giovannoni. Phylogenetic analysis of coastal marine bacterioplankton by 16S rRNA gene cloning from a natural plankton population. Talk presented at the 1994 Ocean Sciences Meeting, San Diego, California, USA, 21-25 February 1994.

34. **Rappé, M. S.**, N. L. Fowles, and S. J. Giovannoni. Identification of a novel microbial 16S rRNA lineage of the Chloroflexus/Herpetosiphon phylum in Sargasso Sea bacterioplankton. Poster presented at the Northwest Branch Meeting of the American Society for Microbiology, Pullman, Washington, USA, 17-19 June 1993.

35. **Rappé, M. S.**, N. L. Fowles, and S. J. Giovannoni. Identification of a novel microbial 16S rRNA lineage of the Chloroflexus/Herpetosiphon phylum in Sargasso Sea bacterioplankton. Poster presented at the ASLO, SWS Annual Meeting, Edmonton, Alberta, Canada, 30 May - 3 June 1993.

APPENDIX 1

State of Hawai'i
DLNR
Northwestern Hawaiian Islands State Marine Refuge
Permit Application Form

For Office Use Only
Permit No:
Expiration date:
Date Appl. Received: N/A
Appl. Fee received: 3/8/06
NWHI Permit Review Committee date:
Board Hearing date:
Post to web date:

Type of Permit

☑ I am applying for a **Research, Monitoring & Education** permit. (Complete and mail Application)
- ☑ This application is for a NEW project in the State Marine Refuge.
- ☐ This application is for an ANNUAL RENEWAL of a previously permitted project in the State Marine Refuge.

☐ I am applying for a permit for a **Native Hawaiian** permit. (Complete and mail Application)
- ☐ This application is for a NEW project in the State Marine Refuge.
- ☐ This application is for an ANNUAL RENEWAL of a previously permitted project in the State Marine Refuge.

☐ I am applying for a **Special Activity** permit. (Complete and mail Application)
- ☐ This application is for a NEW project in the State Marine Refuge.
- ☐ This application is for an ANNUAL RENEWAL of a previously permitted project in the State Marine Refuge.

Briefly describe **Special permit** activity:

When will the NWHI activity take place?

☑ **Summer** (May-July of 2006 (year)
Note: Permit request must be received before February 1st
Specific dates of expedition 18th May - 14th June

☐ **Fall** (August-November) of ____ (year)
Note: Permit request must be received before May 1st
Specific dates of expedition _____

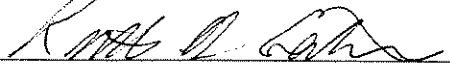
☐ **Other**

NOTE: INCOMPLETE APPLICATIONS WILL NOT BE ACCEPTED

Please Send Permit Applications to:

NWHI State Marine Refuge Permit Coordinator
State of Hawai'i
Department of Land and Natural Resources
Division of Aquatic Resources
1151 Punchbowl Street, Room 330
Honolulu, Hawai'i 96813

NWHI State Marine Refuge Permit Application
See Appendix 2 for Application Instructions

Section A – Applicant Information	
1. Project Leader (attach Project Leader's CV or resume) ☑ CV attached Gates, Ruth, D Name: Last, First, Middle Initial	Dr Title
2. Mailing Address (Street/PO Box, City, State, Zip) Hawaii Institute of Marine Biology P.O. Box 1346 Kaneohe, HI 96744	Telephone (808) 236 7420 Fax (808) 236 7443 Email Address rgates@hawaii.edu
3. Affiliation (Institution/Agency/Organization) Hawaii Institute of Marine Biology/SOEST, University of Hawaii	For graduate students, Major Professor 's Name & Telephone N/A
4. Sub-Permittee/Assistant Names, Affiliations, and Contact Information ☑ CV or resume attached Michael Stat (address as above) Benjamin R Wheeler II (address as above)	
5. Project Title Identifying robust biological indicators of coral disease and/or bleaching susceptibility	
6. Applicant Signature 	7. Date (mm/dd/yyyy) 03/08/2006

Section B: Project Information

8. **(a) Project Location**

☑ NWHI State Marine Refuge (0-3 miles) waters surrounding:

- ☑ Nihoa Island
- ☐ Necker Island (Mokumanamana)
- ☑ French Frigate Shoals
- ☐ Laysan
- ☐ Maro
- ☑ Gardner Pinnacles
- ☐ Lisianski Island, Neva Shoal
- ☐ Pearl and Hermes Atoll
- ☐ Kure Atoll, State Wildlife Refuge
- ☐ Other NWHI location

Describe project location (include names, GPS coordinates, habitats, depths and attach maps, etc. as appropriate).

see attached

(b) check all actions to be authorized:

☑ Enter the NWHI Marine Refuge waters

☑ Take (harvest) ☑ Possess ☑ Transport (☑ Inter-island ☐ Out-of-state)

☐ Catch ☐ Kill ☐ Disturb ☐ Observe

☐ Anchor ☐ Land (go ashore) ☐ Archaeological research

☐ Interactions with Sea Turtles or Monk Seals ☐ Interactions with Seabirds

☑ Interactions with Live Coral, Ark Shells or Pearl Oysters

☐ Interactions with Jacks, Grouper or Sharks

☐ Conduct Native Hawaiian religious and/or cultural activities

☐ Other activities _____

(c) Collection of specimens – collecting activities (would apply to any activity):

Organisms or objects (List of species, if applicable, add additional sheets if necessary):

Common name **Scientific name** **No. & size of specimens** **Collection Location(s)**

see Appendix 1

(d) What will be done with the specimens after the project has ended?

DNA will be extracted and archived at HIMB. Samples will be consumed by the process. If this application is approved, then a subset will be shared with Stephen Karl, Michael Rappe and Rob Toonen at HIMB who have permits pending for the same expedition.

(e) Will the organisms be kept alive after collection? ☐ yes ☑ no

- Specific site/location _____
- Is it an open or closed system? ☐ open ☐ closed
- Is there an outfall? ☐ yes ☐ no
- Will these organisms be housed with other organisms? If so, what are the other organisms?

(Please attach additional documentation as needed to complete the questions listed below)

9. Purpose/Need/Scope:

- See attached section 9

Describe how your proposed activities will help provide information or resources to fulfill the State Marine Refuge purpose and to reach the Refuge goals and objectives.

- See attached section 9

- Describe context of this activity, include history of the science for these questions and background.

  See attached section 9

- Explain the need for this activity and how it will help to enhance survival or recovery of refuge wildlife and habitats.

  See attached section 9

- Describe how your proposed project can help to better manage the State Marine Refuge.

  See attached section 9

10. Procedures (include equipment/materials)

See attached section 10

11. Funding sources (attach copies budget & funding sources).

Funding source attached - NMSP MOA 2005-008/66882

12. List all literature cited in this application as well as all other publications relevant to the proposed project.

See attached section 12

13. What types of insurance do you have in place? (attach documentation)

☐ Wreck Removal

☐ Pollution

14. What certifications/inspections do you have scheduled for your vessel? (attach documentation)

☑ Rat free ☐ tender vessel ☐ gear/equipment

☑ Hull inspection ☑ ballast water

15. Other permits (list and attach documentation of all other required Federal or State permits).

Pending permits to Fish and Wildlife Service and NWHI Coral Reef Ecosystem Reserve

16. Project's relationship to other research projects within the NWHI State Marine Refuge, National Wildlife Refuge, NWHI Coral Reef Ecosystem Reserve, or elsewhere.

National Marine Sanctuary program and HIMB Partnership. NMSP MOA 2005-008/66882

Section C: Logistics

17. Time Frame:

Project Start Date	Project Completion Date
05/01/2006	04/30/2007

Dates actively inside the State Marine Refuge.

May/June 2006 26 days; September 2006

Personnel schedule in the State Marine Refuge (describe who will be where and when).

see attached 17

18. Gear and Materials

- [x] Dive equipment
- [ ] Radio Isotopes
- [x] Collecting Equipment
- [x] Chemicals (specify types)

Ethyl Alcohol (MSDS attached)

19. Fixed installations and instrumentation.

- [ ] Transect markers
- [ ] Acoustic receivers
- [ ] Other (specify)

20. Provide a time line for sample analysis, data analysis, write-up and publication of information.

see attached 20

21. **Vessel Information**:

Vessel Name HI'IALAKAI
IMO Number 8835619
Vessel Owner US Dep. Com. NOAA
Flag USA
Captain's Name CDR Scott Kuester
Chief Scientist or Project Leader
Vessel Type Ocean. Res.
Call sign WTEY
Length 224'
Gross tonnage 1,914
Port of Embarkation
Last port vessel will have been at prior to this embarkation
Total Ballast Water Capacity: Volume see attac$_{m3}$
Total number of tanks on ship 10
Total Fuel Capacity: 228,642 @98%
Total number of fuel tanks on ship 15
Other fuel/chemicals to be carried on board and amounts:

see Appendix 2

Number of tenders/skiffs aboard and specific type of motors:

see Appendix 2

Does the vessel have the capability to hold sewage and grey-water? Describe in detail.

see Appendix 2

Does the vessel have a night-time light protocol for use in the NWHI? Describe in detail (attach additional pages as necessary)

see Appendix 2

On what workboats (tenders) will personnel, gear and materials be transported within the State Marine Refuge?

see Appendix 2

How will personnel, gear and materials be transported between ship and shore?

see Appendix 2

If applicable, how will personnel be transported between islands within any one atoll?

see Appendix 2

Project Location
Coral wil be sampled at sites with the following coordinates:
Nihoa: 23 03 39 North; 161 56 07 West
French Frigate Shoals: 23 43 51 N; 166 09 54 W
Gardner Pinnacles: 25 21 58 N; 170 31 09 W
Johnston Atoll: 16 44 31 N; 169 27 32 W

Purpose/Need/Scope Section 9
The purpose of this research is to identify robust biological indicators of coral disease and/or bleaching susceptibility. Specifically, we will characterize the diversity of symbiotic dinoflagellates (*Symbiodinium*) harbored by corals and examine morphological traits such as polyp size, colony size and colony morphotype in healthy, diseased and/or bleached corals to identify biological traits that correlate with health state. In addition, we will evaluate the diversity of *Symbiodinium* in corals located in the Northwestern Hawaiian Islands and free living in the surrounding waters to examine the prevalence and geographic spread of *Symbiodinium* that render corals disease and/or bleach susceptible in the NWHI.

We hypothesise that
1) Specific coral-*Symbiodinium* assemblages render a coral more susceptible to disease and/or bleaching in the NWHI
2) A suite of morphological traits in corals correlate with disease and/or bleach susceptibility in the NWHI
3) A high diversity of *Symbiodinium* exist in the corals and seawater surrounding the reefs in the NWHI

Our understanding of the biological factors that govern disease and/or bleaching susceptibility is very poor. Our previous work conducted on samples collected during September 2005, shows that diseased *Acropora cytherea* contain a specific type of symbiotic dinoflagellate that is quite different from the type found in their healthy counterparts. The type of symbiont found in the diseased corals is extremely rare in the Pacific and given the implications for coral health, it is critical that we obtain an understanding of the prevalence and geographic spread of this rare symbiont in the NWHI. The work covered by this permit request specifically addresses this need by examining the symbiont types harbored by a range of healthy and diseased and/or bleached corals across the NWHI and examining the abundance of this and other types of algal symbionts in the waters surrounding reefs in the NWHI. In addition we will test the hypothesis that morphological traits that can be non destructively evaluated are informative with regard to the disease and/or bleaching susceptibility of a coral with a view to using these traits for monitoring the health of corals on the NWHI and elsewhere in the future. In total, this research will provide managers with information regarding the geographic spread of symbionts that render corals susceptible to disease and/or bleaching in the NWHI; allow managers to make predictions regarding the susceptibility of given reefs to disease and/or bleaching; develop tools for monitoring coral health and thus build scientific knowledge and capacity that will ultimately reflect in the more effective management of the NWHI coral reef reserve.

The scleractinian corals provide the structural and biological framework that supports the high diversity of marine organisms that inhabit coral reef ecosystems. As such, the health status and functional integrity of coral has profound ramifications for other members of these environments (Hoegh-Guldberg 1999). Corals are susceptible to a variety of environmental disturbances that include changes in sea water temperature, salinity, UV light, pollution, and increased sedimentation (Brown 1997, Williams & Bunkley-Williams 1990, Barber et al 2001, Hoegh-Guldberg 1999). These disturbances are predicted to

increase in frequency due to changes in the global climate and increasing anthropogenic pressure that have implications for even the most remote coral reef ecosystems (Hoegh-Guldberg 1999, Bellwood et al 2004). Coral respond to these environmental insults by losing their symbionts (bleaching) and/ or by exhibiting an increased susceptibility to coral diseases. Ultimately these compromised biological states culminate in reduced growth and reproduction and ultimately, the death of the coral and the degradation of the habitat (Hoegh-Guldberg & Smith 1989, Jokiel & Coles 1977, Gleason & Wellington 1993, Goreau TF 1964, Kushmaro et al 1996, Glynn 1993). One of the most striking facets of corals that are exposed to deleterious conditions or exposed to disease agents is that they do not respond uniformly, that is, different species are differentially sensitive and members of the same species are not equally impacted or susceptible. At this point, our understanding of the biological factors that drive this variation in response are not well developed although we do know that corals form intimate intracellular relationships with a variety of dinoflagellates symbionts, and the taxonomic specifics of these unions potentially influence the vulnerability of corals to environmental disturbance and disease causing agents. These symbionts belong to the genus *Symbiodinium*, which is a highly diverse group that show geographic, depth, and host specificity (LaJeunesse 2005). The type of *Symbiodinium* that a coral hosts has been shown to affect the coral growth rate and thermal tolerance (Little et al 2004, Rowan 2004) and in our previous work in the NWHI we have demonstrated that the type of symbiont found in a coral is tightly correlated with disease state. Other biological traits that have been discussed as contributing to the vulnerability of specific coral to environmental disturbance and or disease agents include colony morphology and coral size (Loya et al 2001). Thus, a detailed understanding of morphological characteristics of the corals combined with a thorough characterization of the types of symbionts they harbor has the potential to be extremely informative about the sensitivity of the specific corals and reef assemblages to environmental shifts and disease agents. Corals that have bleached and or suffered from a disease that reflects as a loss of symbionts, as is the case for many coral diseases (Porter 2001, Green & Bruckner 2001, Sutherland et la 2004; Weil 2004), need to be repopulated with symbionts from pools that are assumed to exist in the environment. Due to technological constraints these pools have never been comprehensively examined before and thus the diversity of symbionts and availability in the environment is completely unexplored. We will address this profound knowledge gap by characterizing patterns of symbiont diversity in the waters surrounding coral reef environments across the NWHI.

Defining the biological basis for, and identifying which corals across the NWHI are at risk from disease agents and/or exhibit an increased propensity to bleach is directly relevant to developing and implementing effective management strategies within the reserve. Our work targets these need by examining how the types of algal symbionts and the morphology of corals map onto disease and/or bleaching susceptibility. These traits lie at the heart of the functional integrity and survival of scleractinian reef corals and thus form the biological foundation on which the success and persistence of the other components of coral reef ecosystems rests.

Defining the types of algal symbionts and coral morphologies that are most commonly associated with diseased and/or bleached corals will allow for the identification of robust biological indicators of coral disease and/or bleaching susceptibility. These tools are directly relevant to management in: 1) identifying areas of the reserve that have a high incidence of disease and/or bleaching susceptible individuals; 2) tracking the spread and prevalence of symbiont types that are associated with disease and/or bleaching susceptibility, and 3) providing a detailed baseline against which future monitoring activities can be compared. In addition, these activities are important in identifying tools that will be globally useful for assessing and understanding declines in coral reef health, and more specifically in examining Pacific wide patterns and evaluating how corals within this ocean are connected to one another in space and time.

Procedures Section 10
A detailed list of coral species to be sampled and the collection strategy for the May/June 2006 cruise is provided in Appendix 1.

To satisfy hypothesis 1: A maximum of 30 coral colonies per species (10 healthy, 10 diseased, 10 bleached) will be sampled from each reef site to determine if there is a correlation between disease susceptibility and/or bleaching and the type of *Symbiodinium* the corals harbor. In reality, the number of samples collected at each location in the NWHI will reflect the incidence of disease and/or bleaching, and our past experiences suggest that individuals representing the compromised health states are rarely encountered. For example, on the September 2005 expedition, coral disease was limited to a single host species, *Acropora cytherea*, found at one reef location, French Frigate Shoals. As such, we have designed a sampling strategy that provides the researchers with the flexibility to take advantage of chance encounters with diseased and/or bleached individuals of the target species at each site visited but that translates as a substantial overestimation of the actual number of corals that will be collected.

To satisfy hypothesis 2: All sampled coral colonies will be photographed for further morphological analysis upon return to the Hawaii Institute of Marine Biology.

To satisfy hypothesis 3: A maximum of 5 coral colonies per species, per site will be sampled to determine the diversity of *Symbiodinium* in coral hosts inhabiting the NWHI. In addition five litres of water (5 replicates per site) will be sampled adjacent to the coral reef and analysed for *Symbiodinium* diversity using methodology newly developed in our laboratory.

Using SCUBA, a 2 cm piece of coral will be removed from each coral sampled using a hammer, cork borer, chisel and/or bone and placed in a plastic zip lock bag. Our experience with these sampling methods suggest that the recovery time is rapid and the biological impacts in terms of the sampled corals and surrounding fauna and flora is minimal. Each sample will be divided on the boat and stored in eppendorf tubes in 70% ethanol or frozen. The samples will be stored in the freezer until the ship returns to port and the material is transported to HIMB for DNA extraction and the downstream analysis. In addition to the suite of analysis outlined here, the coral samples will be characterized for microbial diversity by the Rappe lab and for host genotype by the Toonen and Karl labs at the HIMB.

The seawater samples will be collected in 5L plastic bottles, particulates deposited on a filter using a vacuum pump and then stored in the freezer until DNA extraction can be performed at the HIMB.

Training and Experience of Researchers:
Ruth D. Gates, PhD:
Coral Biologist with 20 years experience working with corals. CV attached.

Michael Stat, PhD:
Coral molecular biologist for past 5 years. Experienced in field work and has performed 3 similar sampling endeavours on the Great Barrier Reef Australia. CV attached.

Benjamin R Wheeler II, MS:
Coral molecular biologist for past year, and microbial molecular biologist for past 7 years. Very experienced in cruise ship expeditions and has participated in 5 previous field trips. CV attached.

Literature cited Section 12

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Loya Y, Sakai K, Yamazato K, Nakano Y, Sambali H, van Woesik R. 2001. Coral bleaching: the winners and the losers. Ecology Letters 4 (2): 122-131
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Williams E & Bunkley-Williams. 1990. The worldwide coral reef bleaching cycle and related sources of coral mortality. Atoll Res. Bull. 335: 1-63.

Personnel schedule Section 17

Ruth D Gates: either May/June or September cruise 2006
Michael Stat: both May/June and September cruises
Benjamin R Wheeler II: both May/June and September cruises

Timeline for analysis Section 20

DNA extraction of sample and morphological analysis completed by August (May cruise) and November (Sept cruise) 2006
Analysis of symbiont diversity completed by December 2006
Submission of publications early 2007

Appendix 1. Sampling Strategy and Collection Request for May/June 2006 Cruise

1. Collections to identify and define the types of symbiotic dinoflagellates harbored by healthy, diseased and bleached corals

Corals belonging to the genera *Acropora* and *Pocillopora* have been chosen for this study because both are sensitive to environmental disturbance and bleach more readily than other species. In addition, preliminary data resulting from the 2005 collections revealed that diseased *Acropora cytherea* harbor a rare and different type of symbiont than their healthy counterparts at French Frigate Shoals. The low prevalence of diseased *Acropora cytherea* in 2005 resulted in statistically inadequate sample sizes so we plan to confirm this important finding by attempting to increase our sample sizes to a statistically relevant number using the collections made in 2006. *Porites lobata* and *Montipora capitata* have also been chosen for this study because they are common and widely distributed within the Hawaiian archipelago and throughout the Pacific and both species are being used as models for work focusing on functional aspects of the symbiosis being conducted at HIMB. These species are very good candidates for experimental manipulations to be conducted at HIMB evaluating the biological implications of forming unions with different symbiont types and in understanding the ecological distribution of these symbioses across the NWHI and at Johnston Atoll.

We are requesting a permit to cover the collection of ten corals (a statistically relevant number) representing each species and health state, for the four sites French Frigate Shoals, Nihoa, Gardner and Johnston. In reality, the number of samples collected at each location in the NWHI will reflect the incidence of disease and/or bleaching, and our past experiences suggest that individuals representing the compromised health states are rarely encountered. For example, on the September 2005 expedition, coral disease was limited to a single host species, *Acropora cytherea*, found at one reef location, French Frigate Shoals. As such, we have designed a sampling strategy that provides the researchers the flexibility to take advantage of chance encounters with diseased and/or bleached individuals of the target species at each site visited (Nihoa, Gardner, Johnston and French Frigate Shoals) but that translates as a substantial overestimation of the actual number of corals that will be collected.

Coral Species (n=10 for healthy, bleached and diseased for 4 locations)

Coral Species	**Common Name**
Acropora. cytherea	Table Coral
Acropora nasuta	Branching Staghorn Coral
Acropora paniculata	Fuzzy Table Coral
Pocillopora damicornis	Lace Coral
Pocillopora meandrina	Cauliflower Coral
*Porites lobata**	Lobe Coral
*Montipora capitata**	Rice Coral

* indicates coral samples that will be shared with Dr Stephen A. Karl, HIMB.

Request Total: 840 corals
Estimate of actual collection: < 350 corals

2. Collections to define the diversity and distribution of symbiotic dinoflagellates harbored by corals across the NWHI and Johnston Atoll

This study will examine a greater diversity of coral species (see below) and to complete this work we plan to collect five healthy corals per species at each of four locations, French Frigate Shoals, Nihoa, Johnston and Gardner. Note that the data collected for the 10 healthy *Acropora cytherea, Acropora nasuta, Acropora paniculata, Pocillopora damicornis, Pocillopora meandrina and Montipora capitata* detailed above will be incorporated in the analyses completed with the corals described below.

Coral Species	**Common Name**
Pocillopora eydouxi	Antler Coral
Porites brighami	Brigham's Coral
Porites lichen	Lichen Coral
Montipora patula	Sandpaper Rice Coral
Leptastrea bewickensis	Bewick Coral
Pavona varians	Corrugated Coral
Fungia scutaria	Oval Mushroom Coral

Request Total: 140 corals
Estimate of actual collection: 140 corals

Appendix 2

13. What types of insurance do you have in place?

NOAA Ship HI`IALAKAI is a U.S. Government-owned and –operated research vessel and is self-insured by the U.S. Government.

14. What certifications/inspections do you have scheduled for your vessel?

- Rat Free (scheduled with U.S. Dept. of Health and Human Services for April 2006)
- Hull Inspection (scheduled with Hawaii Institute of Marine Biology biolobigsts (normally Scott Godwin) prior to projects working in the Northwestern Hawaiian Islands (NWHI)) to ensure no nuisance algae or other fouling species are transported to the NWHI.
- Ballast water information is transmitted to USCG as required by CFR Title 33, Vol. 2, Parts 151.1500 to 199; IMO Resolution A.868(20); and USCG COMDTPUB P16700.4

21. Vessel Information

Vessel Name – NOAA Ship HI`IALAKAI
IMO Number – 8835619
Vessel Owner – U.S. Dept. of Commerce, National Oceanic and Atmospheric Administration (NOAA)
Flag – USA
Captain's Name – CDR Scott Kuester, NOAA
Chief Scientist or Project Leader – Randall Kosaki, Ph.D., NOAA
Vessel Type – Oceanographic Research
Call Sign – WTEY
Length – 224 feet
Gross Tonnage – 1,914
Port of Embarkation – Honolulu
Last port vessel will have been at prior to this embarkation – Pago Pago, Amer. Samoa
Total Ballast Water Capacity:
Volume – 487 m3 (128,834 U.S. gal.)
Total number of ballast tanks on ship – 10
Total Fuel Capacity:
228,642 U.S. gal. at 98% capacity
Total number of fuel tanks on ship – 15
Other fuel/chemicals to be carried on board and amounts: gasoline – as much as 700 U.S. gal.; lube oil – as much as 10,442 U.S. gal.; numerous other industrial and household chemicals used to operate a 224-foot research vessel
Number of tenders/skiffs aboard and specific type of motors:
Ship's own tenders - 1 each 10 m AMBAR Marine jet boat with Yanmar 370-hp,

Diesel inboard engine
1 each 8 m AMBAR Marine jet boat with Yanmar 315-hp, Diesel inboard engine
2 each 17.5 ft Zodiac inflatable boats, each with one Honda 50-hp, 4-stroke, outboard gasoline engine
1 each 19 ft AMBAR Marine rescue boat with Honda 115-hp, 4-stroke, outboard gasoline engine

Program-provided tenders – 19' Boston Whaler with 135 hp Honda four-stroke outboard

Does the vessel have the capability to hold sewage and grey-water? Describe in detail. The ship has a 4,000 U.S. gal Collection Holding Tank for sewage and grey water. In those waters where effluent may NOT be discharged, sewage and grey water are held in this tank until the ship is in waters where sewage and grey water may be discharged. The ship has a U.S. Coast Guard-approved Marine Sanitation Device (Omnipure model MSD 12 MC) which is used to treat sewage and grey water in those waters where effluent may be discharged.

Does the vessel have a night-time light protocol for use in the NWHI? Describe in detail. Navigation lights are on 24-hours/day. Work lights are put on at night only when conducting CTD operations. Weather decks are not illuminated at night.

On what workboats (tenders) will personnel, gear and materials be transported within the State Marine Refuge? - Personnel, gear and materials may be transported within the State Marine Refuge by the ship or any of the 5 ship's small boats listed above or by the program-provided small boat listed above.

How will personnel, gear and materials be transported between ship and shore? – Personnel, gear and materials may be transported between ship and shore by any of the 5 ship's small boats listed above or by the program-provided small boat listed above.

If applicable, how will personnel be transported between islands within any one atoll? - Personnel may be transported between islands within any one atoll by any of the 5 ship's small boats listed above or by the program-provided small boat listed above.

RUTH DEBORAH GATES

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808 236 7420 (office); 808 236 7493 (lab)
808 236 7443 (fax); rgates@hawaii.edu

EDUCATION

1990	Doctor of Philosophy. The University of Newcastle upon Tyne, UK Thesis title: Seawater Temperature and Algal Cnidarian Symbiosis Advisor: Dr. Barbara E. Brown
1984	Bachelor of Science in Marine Biology (Honors). The University of Newcastle upon Tyne, UK

PROFESSIONAL EXPERIENCE

2003 -	Assistant Researcher (tenure track), Hawaii Institute of Marine Biology, SOEST, University of Hawaii at Manoa
2002 - 2003	Assistant Researcher, Department of Organismic Biology, Ecology and Evolution, University of California, Los Angeles
1997 - 2001	Postdoctoral Researcher, Department of Organismic Biology, Ecology and Evolution, University of California, Los Angeles, with Dr. David Jacobs. Evolutionary and developmental significance of homeobox genes in basal metazoans
1996 - 1997	Postdoctoral Researcher, Department of Organismic Biology, Ecology and Evolution, University of California, Los Angeles, with Dr. Peggy Fong. Physiological ecology of the invasive macroalga *Sargassum muticum*
1995 - 1996	Training in Molecular Biology, Department of Organismic Biology, Ecology and Evolution, University of California, Los Angeles, with Dr. Jeanne Erickson. Genetic engineering of the chloroplast genes encoding for the D1 polypeptide in *Chlamydomonas reinhardtii*
1990 - 1995	Postdoctoral Scholar, Department of Organismic Biology, Ecology and Evolution, University of California, Los Angeles, with Professor Leonard Muscatine. Mechanisms of regulation and de-stabilization of coral dinoflagellate symbiosis
1988 - 1990	Research Technician, Department of Biological Sciences, Lehigh University, with Drs. Barry Bean, Paul Samallow and Murray Itzkowitz 1) Cell biological investigations in human fertility 2) Genetic variation in swordtail fish determined by RFLP analysis 3) X-inactivation in the Virginia opossum 4) Monogamy and aggression in the Texas cichlid 5) Behavioral ecology of Beaugregory damselfish

1988 - 1989	Project Manager, Department of Biological Sciences, Lehigh University and Discovery Bay Marine Laboratory, Jamaica, with Dr. Murray Itzkowitz. The behavioral ecology of Beaugregory damselfish
1986 - 1987	Technician, Discovery Bay Marine Laboratory, Jamaica. 1) Photographic monitoring of permanent underwater study sites 2) Laboratory maintenance
1985	Research Assistant, Discovery Bay Marine Laboratory, Jamaica with Dr. Mike LaBarbera. The biomechanics of Jamaican brachiopods
1985 - 1987	Doctoral Researcher, Discovery Bay Marine Laboratory, Jamaica

GRANTS - Current

Duration	2004-2007
Direct:	US $343,980 (best estimate)
Indirect:	US $196,020 (best estimate)
PIs:	Birkeland, C.; Baker, A. C.; Garrison, V. H.; Gates, R. D.; Kellogg, C.; Piniak, G.; Rappé, M.; Toonen, R. J.; Stillman, J. and van Woesik, R.
Title:	Extrinsic and intrinsic factors affecting the resilience of corals to climate change and their use in designing marine reserves.
Agency:	USGS
Duration:	2004 – 2009
Direct	US $11 million; $75,000 Sub-grant to Gates
Indirect:	$0 – administered through UQ, Australia
PIs:	Multi-investigator, multi institutional collaboration
Title:	Global Coral Reef Targeted Research.
Agency:	WorldBank.
Duration:	2004 -2009
Direct: US	$4.6 million; $75,000 to Gates
Indirect:	$0 – administered through UCSB and CSUN
Title:	LTER: Long-Term Dynamics of a Coral Reef Ecosystem.
PIs:	Russell Schmitt, Sally Holbrook, Robert Carpenter, Peter Edmunds plus 14 collaborating scientists including myself
Agency:	NSF-LTER
Duration:	2005-2007
Direct:	US $26,138 plus a 2-year RA (direct)
Indirect:	US $9,488
Title:	Developing Non-Invasive Biotechnological Tools to Monitor Coral Health.
PIs:	Ruth Gates and Mike Rappe
Agency:	Hawaii Sea Grant
Duration:	2005-2007
Direct:	$183,805 (2005-6) and $385,718 (2006-7) to Gates Aeby and Rappe

Title:	NWHICRER-HIMB CORAL HEALTH ASSESSMENT PROGRAM
PIs:	Ruth Gates, Greta Aeby and Mike Rappe
Agency:	NOAA
Duration:	2006-2007
Direct:	$68,515
Indirect:	$2,485
Title:	Are invasive species of dinoflagellates that cause harmful algal blooms entering Hawaii in the ballast water of commercial ships?
PI:	Ruth Gates
Agency:	DLNR/DAR Hawaii Invasive species council

GRANTS - Pending

Submitted:	2006 for 2006-2009
Direct:	US $446,707
Indirect:	US $162,155
Title:	A comprehensive survey of the diversity and abundance of dinoflagellates belonging to the genus *Symbiodinium* across the reef environments of Hawaii
PI:	Ruth Gates
Agency:	NSF BIO OCE
Submitted:	2006 for 2006-2008
Direct:	$126,000
Indirect:	$4,235
Title:	Monitoring sediment impacts in corals: a genomic approach
PI:	Ruth Gates
Agency:	Coral Reef Land Based Pollution Local Action Strategy

PUBLICATIONS

Apprill, A. M. and Gates, R. D. Submitted. Recognizing the complexity of coral endosymbiotic communities. *Nature*

van Oppen, M. J. H. Gates R. D. Submitted. Understanding the resilience of reef corals: the roles of molecular biology and genetics. Invited Review. *Molecular Ecology*

Apprill A. M., Bidigare, R. R. and Gates, R. D. In review. Variability of photosynthetic pigments in endosymbiotic corals. *Limnology & Oceanography*

Ridgeway, T and Gates R. D. In press. Why are there so few genetic markers available for coral population analyses? *Symbiosis*

Gleason, D. F.; Edmunds, P. J. and Gates, R. D. 2006. Ultraviolet radiation effects on the behavior and recruitment of larvae from the reef coral *Porites astreoides*. *Marine Biology 148: 503–512.*

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Bebenek, I. G., Gates R. D., Morris, J., Hartenstein, V., Jacobs, D. K. 2004. *sine oculis* in the Basal Metazoa. *Development, Genes and Evolution 214 (7): 342-351*

Edmunds, P. J. and Gates, R. D. 2004. Size-dependent differences in the photophysiology of the reef coral *Porites astreoides*. *Biological Bulletin 206: 61-64*

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Gates, R. D., Hadrys, T.; Arenas-Mena, C. and Jacobs, D. K. 2002. Determining spatial and temporal patterns of developmental gene expression in vertebrates and invertebrates using *in situ* hybridization techniques. In*: Methods and Tools in Biosciences and Medicine: Techniques in Molecular Evolution and Systematics (Eds. DeSalle, R.; Giribet, G. and Wheeler, W.). Birkhauser Pp. 365-399*

Edmunds, P. J. and Gates, R. D. 2002. Normalizing physiological data for scleractinian corals. *Coral Reefs 21 (2): 193-197*

Nallur, R. B.; Gates, R. D.; Ladurner, P.; Jacobs, D. K. and Hartenstein, V. 2002. Neurogenesis in the primitive bilaterian Neochildea I. Normal development and isolation of genes controlling neural fate. *Development, Genes and Evolution 212 (2): 55-69*

Lee, S. E.; Gates, R. D. and Jacobs, D. K. 2001. The isolation of a *Distal-less* gene fragment from two molluscs. *Development, Genes and Evolution 211: 506-508*

Edmunds, P. J.; Gates, R. D. and Gleason, D. F. 2001. The biology of planulae freshly released from the reef coral *Porites astreoides* and their response to temperature disturbances. *Marine Biology 139: 981-989*

Jacobs, D. K.; Wray, C. G.; Wedeen, C. J.; Kostriken, R.; DeSalle, R.; Staton, J. L.; Gates, R. D. and Lindberg D. L. 2000. Molluscan *engrailed* expression, serial organization, and shell evolution. *Evolution and Development 2(6): 340-347*

Gates, R. D. and Edmunds, P. J. 1999. The physiological mechanisms of acclimatization in tropical reef corals. *American Zoologist 39(1): 30-43*

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Lee, S.E.; Gates, R. D and Jacobs, D. K. Can Short Sequences of Conserved Genes be Accurately Assigned an Affiliation? *Development, Genes and Evolution*

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ABSTRACTS (Last 5 years)

Leggat, W and Gates, R. D. (2006). The molecular biology of thermal stress in *Symbiodinium*. Verbal presentation at The Society of Experimental Biology meeting, The University of Kent at Canterbury. 2nd - 7th April 2006

Wheeler II, B. R., Stat, M. and Gates, R. D. (2006) Validating the Molecular Markers Used to Infer Phylogeny in the Endosymbiotic Dinoflagellate Genus, *Symbiodinium*, Using a Single Cell Approach. Poster presentation at The 13th Ocean Sciences Meeting 20-24 February 2006 in Honolulu, Hawaii

Stat, M. and Gates, R. D. (2006). Diversity of coral endosymbionts (*Symbiodinium*) across the Northwestern Hawaiian Islands. Verbal presentation at The 13th Ocean Sciences Meeting 20-24 February 2006 in Honolulu, Hawaii

Aprill, A. M. and Gates, R. D. (2006). Assessing the Diversity of Endosymbiotic Dinoflagellates in Hawaiian Corals. Verbal presentation at The 13th Ocean Sciences Meeting 20-24 February 2006 in Honolulu, Hawaii

Manning, M and Gates, R. D. (2006). The Diversity of Dinoflagellates Living in the Waters and Substrates Surrounding the Coral Reefs of Kane'ohe Bay. Verbal presentation at The 13th Ocean Sciences Meeting 20-24 February 2006 in Honolulu, Hawaii

Pagarigan, L., Takabayashi, M. and Gates, R. D. (2006). Assessing Environmentally Induced Stress in Scleractinian Corals at the Molecular Level. Poster presentation at The 13th Ocean Sciences Meeting 20-24 February 2006 in Honolulu, Hawaii

Shannon, T.; Gates, R. D. and Fitt, W. K. 2003. Algal symbioses and the potential for osmotic stress: how do symbiotic reef invertebrates deal with it? 7th International Conference on Ceolenterate Biology, Lawrence, Kansas.

Nallur, R. B.; Gates, R. D.; Jacobs, D. K.; Ladurner, P.; Reiger, R. and Hartenstein, V. 2001. Early neurogenesis in flatworms. *Belgian Journal of Zoology. 131(Supplement 1). April. 67.*

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Gates, R. D. and Edmunds, P. J. 2001. Osmotic Stress: A potential biomarker for monitoring coral bleaching. *UNESCO/IOC Paris.*

Jacobs, D. K.; Gates, R. D. 2001. Evolution of POU/homeodomains in basal metazoa: Implications for the evolution of sensory systems and the pituitary. *(Sixtieth Annual Meeting of the Society for Developmental Biology Seatle, WA, USA July 18-22, 2001). Developmental Biology 235 (1): 241.*

Jacobs, D. K., Gates, R. D. and Palchevskiy, V. A. 2000. Sponge muscles, cnidarian brains, and how Onychophora lost their stripes: Developmental genetics, and the metazoan radiation. *West Coast Regional Developmental Biology Conference, Bodega Marine Laboratory.*

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Nallur, R. B.; Mazzotta, J.; Dumstrei, K.; Gates, R. D.; Lee, S. E.; Jacobs, D. K. and Hartenstein, V. 2000. Role of *dpp/sog* and the homeobox genes *vnd, ind,* and *msh* in brain development. *International Fruit Fly Conference.*

INVITED PRESENTATIONS

2005	18th International EPSCoR Conference, Puerto Rico
2005	Committee for the Coral Reef Land-Based Pollution LAS, EPA
2005	World Bank Meeting, Puerto Morales, Mexico
2005	Department of Biology, California State University, Northridge, CA
2005	Department of Oceanography, University of Hawaii, HI
2004	Marine Biotechnology Outreach and Education Initiative, HIMB, HI
2003	Public Lecture, UCLA Astrobiology Institute
2002	Department of Zoology, University of Hawaii, HI
2002	Department of Zoology, Oregon State University, Corvallis, OR
2001	Department of Biology, Lehigh University, PA
2001	Department of Biology, California State University, Northridge, CA
1998	Bodega Bay Marine Laboratory, CA
1998	Society for Integrative and Comparative Biology, Boston, MA
1996	Department of Biology, California State University, Northridge, CA
1994	Office of Naval Research, contractors meeting, San Francisco, CA
1991	Office of Naval Research, contractors meeting, Maui, HI
1991	Department of Biology, UCLA, CA
1991	Scripps Institute of Oceanography, University of California, San Diego, CA

EDUCATIONAL ACTIVITIES

Postdoctoral Advisee

Michael Stat (2005 – 2007)

Graduate Student mentor and Committee Chair

Craig Coleman (PhD candidate) Department of Oceanography, UHM
Marissa Hirst (PhD candidate) Department of Zoology, UHM
Jackie Padilla Gamino (PhD candidate) Department of Oceanography, UHM
Mackenzie Manning (MS candidate) Department of Zoology, UHM
Anderson Mayfield (PhD candidate) Department of Zoology, UHM
Dan Reineman (PhD candidate) Department of Oceanography, UHM

Additional Graduate Committees

Dan Barshis (PhD candidate) Department of Zoology, UHM
Brian Boeing (MS candidate) Department of Oceanography, UHM
Stuart Ibsen (PhD candidate) Department of Zoology, UHM
Mahealani Kaneshiro (MS candidate) Department of Zoology, UHM
Emily Morris (MS Candidate) Humboldt State University, US
Lauren Pagarigan (MS Candidate) Marine Science Department, UHH